

A MATHEMATICAL-STATISTICAL MODEL FOR THE ANALYSIS OF CROSS-SECTIONAL SEROLOGICAL DATA WITH SPECIAL REFERENCE TO THE EPIDEMIOLOGY OF MALARIA

A METHODOLOGY FOR THE DETECTION OF CHANGE IN THE TREND OF TRANSMISSION



J. A. M. van DRUTEN

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krips repro meppel

TO HETTY, ALRIK AND ARJAN

AND TO MY PARENTS

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CONTENTS

CHAPTER 1	GENERAL INTRODUCTION	1
CHAPTER 2	THE TREND OF TRANSMISSION IN A HOMOGENEOUS POPULATION	
2.1	<i>The time-dependent transmission model</i>	5
2.1.1	Introduction	5
2.1.2	Transmission assumptions	7
2.1.3	The transmission intensity	8
2.1.4	The rate of change in transmission intensity	12
2.1.5	The trend of transmission intensity	12
2.1.6	The rate of change in the trend of transmission intensity	15
2.1.7	Statistical inference, the trend-change test	16
2.2	<i>Illustration; sero-epidemiological data from Mauritius</i>	19
2.3	<i>Correction for age specific malaria death rate and other causes of death</i>	26
2.4	<i>Discussion</i>	29
2.4.1	General remarks regarding methodology	29
2.4.2	The concept of inoculation	31
2.4.3	The transmission assumptions	34
2.4.4	The transmission function, explanatory variables	35
2.4.5	Spline approximations	36
2.4.6	The p-value function	38
2.4.7	Comments on the application of the model	39
2.5	<i>Summary</i>	43

CHAPTER 3 THE TREND OF TRANSMISSION IN A NON-HOMOGENEOUS POPULATION	
3.1 <i>Introduction</i>	45
3.2 <i>Comparison of the trend in transmission intensity of two homogeneous populations</i>	46
3.3 <i>Spatial-dependent and time-dependent transmission model</i>	49
3.3.1 <i>Introduction</i>	49
3.3.2 <i>Parameters for a non-homogeneous population</i>	50
3.3.3 <i>Statistical inference, trend-change test</i>	60
3.3.3.1 <i>Introduction</i>	60
3.3.3.2 <i>Fixed design</i>	62
3.3.3.3 <i>Random design</i>	67
3.3.3.4 <i>Relationship between the analyses based on an F-design and on an R-design</i>	70
3.4 <i>Illustration, using data collected in Sri Lanka</i>	75
3.4.1 <i>Introduction</i>	75
3.4.2 <i>The design of the survey</i>	76
3.4.3 <i>Analysis of serological data</i>	78
3.4.4 <i>Analysis of parasitological data for the years 1973-1978</i>	89
3.5 <i>Discussion</i>	97
3.5.1 <i>Remarks regarding methodology</i>	97
3.5.2 <i>Comparison of the spline trend $\hat{\lambda}_s(t)$ with the mean SPR trend</i>	102
3.5.3 <i>Comparison of the spline trend $\hat{\lambda}_s(t)$ with the mean SPR trend at sub-area level</i>	102
3.5.4 <i>Indications for the effectiveness of malathion</i>	104
3.5.5 <i>Spatial variation</i>	104
3.5.6 <i>Further research</i>	107
3.6 <i>Summary</i>	107
CHAPTER 4 CORRECTION FOR FADING OF ANTIBODIES AND SENSITIVITY AND SPECIFICITY OF THE SEROLOGICAL TEST	
4.1 <i>Introduction</i>	109
4.2 <i>The mathematical model</i>	110
4.2.1 <i>Introduction</i>	110
4.2.2 <i>Past-sensitivity η and past-specificity ξ of a serological test</i>	112

4.2.3 Instantaneous correction for the factor past-sensitivity	120
4.2.3.1 Introduction	120
4.2.3.2 The sign disturbance coefficient	121
4.2.3.3 Past-sensitivity is independent of age, is constant and its level is unknown	124
4.2.3.4 Past-sensitivity changes with age; linear correction	126
4.2.3.5 Interpretation of the course of the antibody related transmission function	134
4.2.4 Detection of a downward trend of the transmission intensity in a homogeneous population	137
4.3 Illustration; sero-epidemiological data from Mauritius	139
4.4 Extensions of the model; correction for the simultaneous effect of the past-sensitivity and past-specificity of the serological test	144
4.5 General discussion	147
4.5.1 The past-specificity of the test	148
4.5.2 Relationship between the course of the spline approximations $\lambda_s^a(t)$ and $\lambda_s(t)$	149
4.5.3 The curvature of the past-sensitivity	151
4.5.4 The validity of the two-step procedure	152
4.5.5 The critical past-sensitivity	154
4.5.6 Comments on the application	156
4.6 Summary	157
CHAPTER 5 APPLICATIONS	
5.1 General introduction	159
5.2 Sero-epidemiological investigations of malaria in Guyana	160
5.2.1 Introduction	160
5.2.2 The design of the study	160
5.2.3 Results	161
5.2.4 Discussion	165
5.3 Malaria in Surinam, a sero-epidemiological study	168
5.3.1 Introduction	168

5.3.2 Results of the analysis using the constant infection rate model	169
5.3.3 Application of the two-step procedure	169
5.3.4 Discussion	176
5.4 Serological data collection in Sri Lanka	184
5.4.1 Introduction	184
5.4.2 Trend of the transmission intensity in the sub-areas	185
5.4.3 Assessment of the overall trend	189
5.4.4 Discussion	192
5.5 Nigeria, Garki-project	197
5.5.1 Introduction	197
5.5.2 Serological data and study design	198
5.5.3 Detection of a downward trend in the transmission intensity	199
5.5.4 Discussion	203
5.6 Serological data from Panama	205
5.6.1 Introduction	205
5.6.2 The trend of the transmission intensity in Chepigana and Pinogana	206
5.6.3 Assessment of the overall trend	212
5.6.4 Trend based on seroreactivity to <i>P.vivax</i> and <i>P.falciparum</i>	216
5.6.5 Discussion	216
5.7 Summary	226
CHAPTER 6 GENERAL DISCUSSION	
6.1 The present malaria model in relation to other mathematical approaches	229
6.2 Applicability of the model to other fields of research	235

APPENDIX 1

EPIDEMIOLOGICAL PARAMETERS AND NOTATIONAL CONVENTIONS

1.1 Parameters for the description of the trend in transmission intensity	239
1.2 Age related parameters	241
1.3 Antibody related transmission functions	242

1.4	Notation of estimates of parameters, description of time and age	245
1.5	General legend for the interpretation of the curve $\lambda_s^a(t)$, the spline approximation	247

APPENDIX 2

THE TREND OF TRANSMISSION IN A HOMOGENEOUS POPULATION

2.1	The time-dependent transmission model	249
2.1.1	The non-stationary Poisson process	249
2.1.2	Spline approximations	252
2.2	Statistical inference	257
2.2.1	Determination of the functions $l_j(t)$, $c_j(t)$; $j=0, 1, \dots, k$	258
2.2.2	Estimation and testing procedures	265

APPENDIX 3

THE TREND OF TRANSMISSION IN A NON-HOMOGENEOUS POPULATION

3.1	Spatial-dependent and time-dependent transmission model	275
3.2	Statistical inference	280
3.2.1	Fixed design	281
3.2.2	Random design	286
3.2.3	Relationship between the analyses based on an F-design and on an R-design	293

APPENDIX 4

CORRECTION FOR FADING OF ANTIBODIES AND SENSITIVITY AND SPECIFICITY OF THE SEROLOGICAL TEST

4.1	Introduction	297
4.2	Past-sensitivity and past-specificity and their relationship with sensitivity, specificity and fading of antibodies	298
4.3	Interpretation of the antibody related transmission function $\lambda^d(t)$	302
4.3.1	Relationship between $\lambda(t)$ and $\lambda^d(t)$	302
4.3.2	The sign disturbance coefficient $\Lambda(A)$	305
4.3.3	The critical past-sensitivity $\eta_c(A)$	308
4.3.4	The sign of $\gamma(A)$	310

4.3.5 Maximum linear correction (MLC) method for the factor of past-sensitivity	315
4.4 Extensions and generalizations	317
4.4.1 Constant past-sensitivity η and constant past-specificity ξ	317
4.4.2 Variable past-sensitivity $\eta(A)$ and variable past-specificity $\xi(A)$	318
4.4.2.1 Basic theorem	319
4.4.2.2 Relationship between $\lambda^a(t)$ and $\lambda(t)$	325
4.4.2.3 Relationship between $\lambda_s^a(t)$ and $\lambda_s(t)$	327
4.4.2.4 Relationship between $\lambda_s^a(t)$ and $\lambda_s(t)$	329
<i>References</i>	333
<i>Author index</i>	337

GENERAL INTRODUCTION

1

In this study an attempt is made to develop a mathematical model for the assessment of the trend of malaria transmission by using cross-sectional serological data.

Normally, the presence of malarial antibodies in the serum is the result of either an actual or a past malaria parasitaemia. A number of serological tests developed to detect and measure antibodies specific for the blood stages of the parasites are currently used for epidemiological studies of the population. Following the introduction of the IFA test in 1961 (*Tobie et al.*), the potential importance of serological surveys for the assessment of the trend of malaria transmission was underlined in various papers and in recommendations of advisory groups of the *World Health Organization* (1968, 1975). However, the mathematical-statistical methodology for the analysis of cross-sectional serological data had not yet been sufficiently developed. This was probably due to the complexity of the process involved and the simultaneously

interacting variables which determine the serological profile of the community.

The validity of a model depends almost entirely on whether the factors and the associated assumptions included in the model, turn out to be those which are really essential for the explanation of the observed data.

Muench (1959) states: "Biological entities are so complex that it is impossible ever to choose more than a small fraction of all the factors which might be of importance. Clearly, a great deal must always be neglected in any mathematical study". This applies also to models for the study of the epidemiology of malaria. The model presented here takes into account the following factors:

F_1 : the time-dependent risk of receiving inoculations

F_2 : the spatial-dependent risk of receiving inoculations

F_3 : the immune response of the individuals

F_4 : the sensitivity of the serological test

F_5 : the specificity of the serological test.

Assumptions are needed for simplifying the basic factors F_1 to F_5 to establish the mathematical-statistical relations between these factors and the observed serological data. Three major problems to be solved were:

A. The construction of a time-dependent transmission model for the description of varying transmission, which should also include parameters to describe the trend of transmission (factor F_1).

B. The incorporation into the model of the spatial variation in the trend of the transmission intensity (factor F_2).

C. The introduction into the model of corrective measures for the factors fading of antibodies, the sensitivity and specificity of the serological test (factors F_3 , F_4 , F_5).

An attempt is made to solve these problems one by one in the chapters 2, 3 and 4 respectively.

The time-dependent transmission model presented in chapter 2 uses a spline approximation of the intensity function of a non-stationary Poisson process as a measure of the trend of transmission in a homogeneous population. The model provides a general parametrisation for the time-dependent force of infection in the catalytic epidemic model of Muench (1953). Although the model presented here is in relation to the epidemiology of malaria and the analysis of data from cross-sectional serological surveys, its applications are not restricted to the above only. The frame-work of the model is general enough to be applicable to other infectious diseases especially for the analysis of data from cohort studies.

Chapter 3 is concerned with the problem of determining the trend of transmission in a non-homogeneous population. Here the spatial variation in the risk of receiving an inoculum is additionally implemented.

Chapter 4 deals with the most difficult problem viz. to take into account the normally occurring decrease of seroreactivity in the absence of a continuing antigenic stimulation as well as the sensitivity and the specificity of the serological test. Lack of attention to these factors will of course result in biased estimates of the level and trend in transmission intensity. Many factors affect an individual's immune response to malaria. Among these are his age and immuno-competence, his cumulative time-related exposure to malaria antigens and the kind and amount of specific therapy which he may have received. Reliable quantitative data regarding these are generally not available. These factors are therefore unsuitable for a direct quantitative assessment and incorporation into the model. Concepts which merge several factors into one are therefore employed. For that reason the concepts of "past-sensitivity" and "past-specificity" of a serological test are introduced. The past-sensitivity of a test measures the capability of the test to detect either current or past malaria parasitaemias in individuals of the population

and depends on the fading of antibodies (factor F_3) as well as on the sensitivity and the specificity of the serological test (factors F_4 and F_5).

Chapter 4 uses the "biased" transmission functions introduced in chapter 2, but additionally corrects for any level and linear change of the unknown past-sensitivity by age, while assuming that the past-specificity of the test is 1. Using this type of correction - called *maximal linear correction (MLC)* for the factor of past-sensitivity - an indirect standardized method is developed for the detection of a downward trend in transmission intensity.

In chapter 5 serological data from Mauritius, Guyana, Surinam, Nigeria, Panama and Sri Lanka are used to illustrate the application of the theory. In the applications a standard way of presentation i.e. a standard figure is used to explain the successive steps in the detection of a change in the trend of transmission. The general legenda for these standard figures is presented in Appendix 1 (section 1.5). This appendix also provides a comprehensive list of epidemiological parameters. The conventions regarding the notation of these parameters, their estimates (estimators) with standard deviations are also explained in Appendix 1.

The mathematical-statistical elements of the models introduced in chapter 2, 3 and 4 are presented in detail in Appendix 2, 3 and 4 respectively. These three appendices are to a large event self-explanatory and the mathematically skilled reader can use these without a frequent consultation of the corresponding chapters.

THE TREND OF TRANSMISSION IN A HOMOGENEOUS POPULATION

2

2.1 THE TIME-DEPENDENT TRANSMISSION MODEL

2.1.1 Introduction

During the past 70 years most of the work on mathematical models in the epidemiology and control of tropical diseases was devoted to malaria. The mathematical approach to the control of malaria goes back to the work of Ross (1911) "Prevention of Malaria" under the title "The theory of happenings". The value of mathematical techniques in the epidemiology and control of malaria has been studied with renewed interest during the past two decades, since the publication of Macdonald's book in 1957. Macdonald's approach stresses the entomological aspects of malaria transmission. Basic epidemiological parameters in his model are the inoculation rate and the basic reproduction rate. The basic reproduction rate has been adapted by Garrett-Jones (1964) and his concept of vectorial capacity combines most of the relevant information about

the mosquito population in a single parameter.

Dietz, Molineaux and Thomas (1974) are responsible for the further developments in malaria modelling. This model, which is based on a long list of assumptions tries to relate the entomological and parasitological observations while taking into account the effect of immunity on transmission. This model was considered epidemiologically satisfactory and fit for use in planning malaria control operations (Dietz et al. 1978). The basic principles of these and other mathematical models of infectious diseases are presented in a book by Bailey (1975). The essentials of malariology, including malaria epidemiology are recently reviewed by Bruce-Chwatt (1980).

After the introduction of serological tests comparatively little attention has been given to the development of models using serological data. In the field of malaria Draper, Voller and Carpenter (1972) introduced a statistical model for the estimation of a constant "infection rate" using the rising seropositivity rate in different age groups. This model is in principle a catalytic model with a constant force of infection (Muensch, 1959). Although the major factors F_1 to F_5 (see chapter 1) are not taken into account, Bruce-Chwatt (1976) considers that it has been successfully applied in East Africa, Mauritius, Surinam and Greece.

The time-dependent transmission model presented here is a first step towards the construction of a new model for the analysis of cross-sectional serological data. It can be considered a generalization of the constant infection rate model of Draper et al. (1972). In analogy with these authors it is presupposed in the present model that there are means to separate in the population examined those individuals who have already experienced a parasitaemia from those who have not. Four transmission assumptions and the concept of inoculation are basic in the model. The intensity function of a

non-stationary Poisson Process is used to describe the time-dependent risk of receiving an inoculation. A spline approximation of this function is used as a measure of the trend in transmission. The model takes into account the statistical fluctuations in the data due to the sampling of individuals and a statistical test ("*trend-change test*") is introduced for the detection of a down- or upward trend in transmission intensity.

2.1.2 Transmission assumptions

An individual of a population is considered to be exposed to a time-dependent risk of receiving infective inocula. The basic random event in time is the occurrence of an inoculation. As will be explained later, the following definition of an inoculation with malaria parasites is used in this study:

An inoculation is the receipt of a quantity of sporozoites from the local mosquito population which is sufficiently infectious to induce a parasitaemia in a previously uninfected individual.

Such an event is called an inoculation irrespective of the fact that it occurs to an individual who already has been infected with sporozoites. For each individual of the population the following assumptions are made on the occurrence of an inoculation:

Transmission assumptions

1. In any time interval, no matter how small, there is a positive probability that an inoculation will occur, although it is not certain that an inoculation will occur.
2. In sufficiently small time intervals we may ignore the probability of two or more inoculations on the same host as compared with the probability of exactly one inoculation.

3. The numbers of inoculations received by an individual in disjunct time intervals are independent in a statistical sense. The probability distributions of the numbers of inoculations in these intervals may vary.
4. The individuals of the population are at time t exposed to the same risk of receiving an inoculum. In this context the population under study is considered to be *homogeneous*.

2.1.3 The transmission intensity

In this section a function $\lambda(t)$ is defined which describes the transmission intensity in a homogeneous population at any time point t before the survey date.

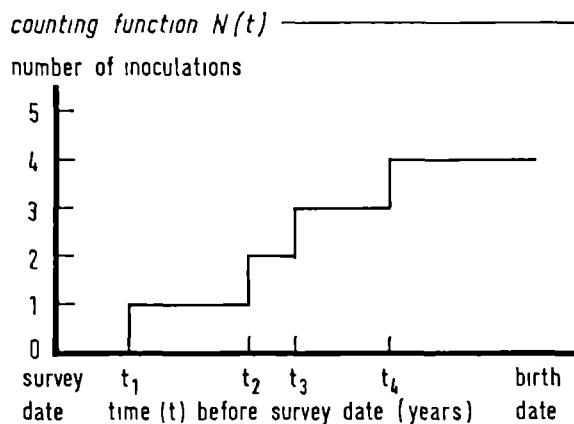
For an individual of age A on survey date the inoculations during the period $[0, A]$ before survey date are the random events under consideration. These events are described by a *counting function* $N(t)$, which represents the number of inoculations that have occurred during the time period between the survey date and a time point t before that date ($0 \leq t \leq A$). For each time t , the value $N(t)$ is a random variable. The collection of random variables $\{N(t); 0 \leq t \leq A\}$ constitutes a stochastic process. A typical realisation of this process is the counting function $N(t)$ shown in fig. 2.1.

On account of partial information, collected by the survey it is supposed that we can distinguish between:

- (i) $N(A) > 0$:the individual has experienced at least one inoculation
- (ii) $N(A) = 0$:the individual has experienced no inoculation.

It can be shown that if the stochastic process $\{N(t); 0 \leq t \leq A\}$ satisfies the transmission assumptions 1, 2 and 3 (see section 2.1.2), then the counting function $N(t)$ follows a Poisson distribution for each t (see

Figure S.1 Graphical representation of a counting function $N(t)$, representing the number of inoculations inflicted on one individual in period $[0, t]$ before survey date; t_1, t_2, t_3, t_4 , denote time points where an inoculation has occurred.



Appendix 2). The expected value of the number of inoculations $N(t)$ in period $[0, t]$ before survey date is denoted by $v(t)$ and is called the *mean value function*,

$$v(t) = \text{mean (expected) number of inoculations in period } [0, t] \quad (2.1)$$

before survey date.

It is assumed that this function is differentiable, with a continuous derivative denoted by $\lambda(t)$,

$$\lambda(t) = \frac{d}{dt} v(t). \quad (2.2)$$

This function will be called the *transmission function* $\lambda(t)$ of the individual under consideration. Mathematically $\lambda(t)$ presents the intensity function of a non-stationary Poisson process. For this application it is

convenient to choose the year as the unit of time. The value of the transmission function $\lambda(t)$ then equals the mean (or expected) number of inoculations per annum inflicted on an individual t years before survey date. There are two additional properties of the transmission function $\lambda(t)$. First the approximate probability that the individual has experienced an inoculation in a small time interval $(t, t + \Delta t)$ before survey date is equal to $\lambda(t)\Delta t$. Second the mean number of inoculations in an arbitrary period $[T_1, T_2]$ before survey date experienced by the individual is presented by the area below $\lambda(t)$ between the time points T_1, T_2 . This is represented by the formula

$$\int_{T_1}^{T_2} \lambda(t)dt = \text{mean number of inoculations in period } [T_1, T_2] \quad (2.3)$$

before survey date.

A graphical illustration of the transmission function $\lambda(t)$ and the integral

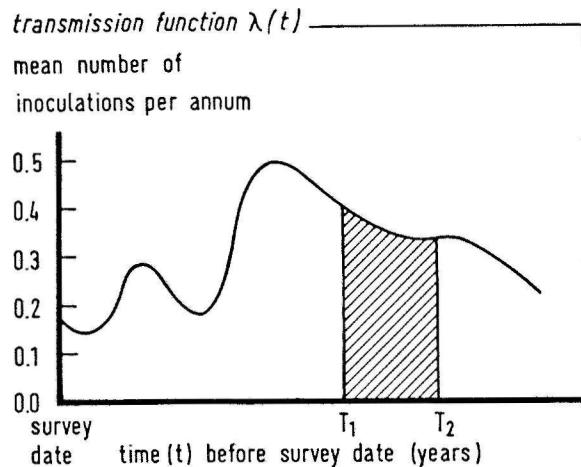
$$\int_{T_1}^{T_2} \lambda(t)dt$$

is presented in fig. 2.2.

So far a transmission function $\lambda(t)$ for each individual of the population has been defined, describing the time-dependent risk of receiving an inoculation from the local mosquito population. Transmission assumption 4 of the model (see section 2.1.2) states that all individuals of the population are exposed at time t before survey date to the same risk of receiving an inoculum. In other words the value of $\lambda(t)$, t years before the survey date, is equal for all individuals of the population under study. This implies that $\lambda(t)$ can be defined as *the transmission function of the population* and that this function can be considered to measure the time-dependent force of infection acting on the population t years before survey date.

As $\lambda(t)$ is the intensity function of a non-stationary Poisson process: the proportion of individuals of age A who have experienced at least one

Figure 2.2 Graphical representation of a transmission function $\lambda(t)$; the shaded surface below $\lambda(t)$ between T_1 and T_2 represents the mean number of inoculations in period $[T_1, T_2]$ before survey date.



inoculation in their life, i.e. the cumulative inoculation rate notated as $p(A)$, is equal to

$$p(A) = 1 - \exp\left(- \int_0^A \lambda(t) dt\right). \quad (2.4)$$

It follows immediately that

$$\frac{dp}{dA} = \lambda(A) (1-p(A)). \quad (2.5)$$

This is exactly the simple catalytic differential equation with a variable infection rate. Hence $\lambda(t)$ can be interpreted as the rate in which previously non infected individuals become infected. Or using the terminology of Muench (1959), $\lambda(t)$ measures the "effective contacts" per year per individual.

2.1.4 The rate of change in transmission intensity

It is assumed that the transmission function $\lambda(t)$ is differentiable with a continuous derivative denoted by $\gamma(t)$,

$$\gamma(t) = \frac{d}{dt} \lambda(t). \quad (2.6)$$

The function $\gamma(t)$ describes the rate of change in the transmission intensity, t years before survey date. The symbol t indicates a time point t years before survey date, so the direction of the conventional time scale has been reversed. Consequently $\gamma(t)$ has the following epidemiological interpretation:

$\gamma(t) > 0$ there has been a decrease in transmission intensity t years before survey date

$\gamma(t) = 0$ there has been no change in transmission intensity t years before survey date

$\gamma(t) < 0$ there has been an increase in transmission intensity t years before survey date.

2.1.5 The trend of transmission intensity

The main objective in this section is the approximation of the transmission function $\lambda(t)$ by a piecewise defined function, i.e. defined separately for successive time intervals, which can be used to describe the trend of the transmission intensity t years before survey date. Such a function is preferred over a single function defined for the entire time period under consideration, as the trend of transmission in one time interval may be totally unrelated to that in another interval. A function defined by one formula for the entire period would have the opposite property: namely its behaviour in a small period would determine its behaviour everywhere. As we shall see later, functions which suit the objective of this study are the so called "mathematical splines". First a brief definition is given of a spline

approximation, notation $\lambda_s(t)$, of the transmission function $\lambda(t)$.

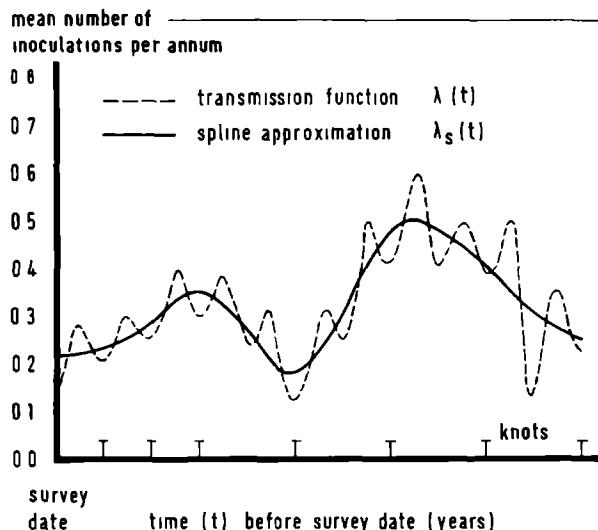
Consider a division of the time axis before survey date in intervals $[0, T_1]$, $[T_1, T_2]$, ..., $[T_{k-1}, T_k]$. The time points $0, T_1, T_2, \dots, T_k$ are called the *knots* of the spline approximation $\lambda_s(t)$. We define the *spline approximation* $\lambda_s(t)$ of the transmission function $\lambda(t)$ by

$$\lambda_s(t) = \frac{d}{dt} v_s(t). \quad (2.7)$$

The function $v_s(t)$ is the so called natural cubic spline approximation of the mean value function $v(t)$ (for further details see Appendix 2).

The spline approximation $\lambda_s(t)$ consists of piecewise defined polynomials of degree 2. The pieces join in the knots and fulfil continuity conditions for the function itself and its derivative. Graphically the spline approximation $\lambda_s(t)$ appears as a flexible continuous curve (see illustration fig. 2.3).

Figure 2.3 Graphical illustration of a spline approximation $\lambda_s(t)$ of the transmission function $\lambda(t)$; the spline function $\lambda_s(t)$ is used to describe the trend of the transmission intensity.



This function $\lambda_s(t)$ is called the 'spline' trend of the transmission intensity.

The function $\lambda_s(t)$ is uniquely defined by the values of the mean value function $v(t)$ in the knots $0, T_1, T_2, \dots, T_k$.

It is convenient to denote these values by v_j , thus

$$v_j = v(T_j) \quad (j=0, 1, \dots, k). \quad (2.8)$$

$$\text{here } T_0 = 0 \text{ and } v_0 = v(T_0) = v(0) = 0.$$

It is possible to express the approximation $\lambda_s(t)$ as a weighted linear combination of the parameters v_1, v_2, \dots, v_k , in formula

$$\lambda_s(t) = l_1(t) v_1 + l_2(t) v_2 + \dots l_k(t) v_k. \quad (2.9)$$

The functions $l_j(t)$ ($j=1, \dots, k$) are quadratic functions of t (see Appendix 2).

The parameters v_j ($j=1, \dots, k$) can be estimated using cross-sectional data (section 2.1.7)

An important property of this spline approximation $\lambda_s(t)$ which suites the present objectives is the following

$$\int_{T_{j-1}}^{T_j} \lambda_s(t) dt = \int_{T_{j-1}}^{T_j} \lambda(t) dt \quad (j=1, \dots, k). \quad (2.10)$$

This property states that the surface of the area below the spline approximation $\lambda_s(t)$ between two adjacent knots is exactly equal to the corresponding surface of the true but unknown transmission function $\lambda(t)$ and is therefore denoted as the *property of preservation of surface*. In other words the surface of the area below the approximation $\lambda_s(t)$ between two adjacent knots presents exactly the mean number of inoculations in this period. Over intervals where $\lambda_s(t)$ is negative (which may occur), the area between the curve and the horizontal axis has to be taken as negative.

2.1.6 The rate of change in the trend of transmission intensity

The main parameter for the evaluation of malaria control operations is the change in the trend of the transmission intensity. The rate of change in the trend function $\lambda_s(t)$ is denoted by $\gamma_s(t)$, thus

$$\gamma_s(t) = \frac{d}{dt} \lambda_s(t). \quad (2.11)$$

The derivative of a spline function is a spline function, so the function $\gamma_s(t)$ can be considered a spline approximation of the rate of change in transmission intensity $\gamma(t)$. Graphically $\gamma_s(t)$ is a broken line, the pieces join in the knots T_1, T_2, \dots, T_{k-1} . The value of the spline approximation $\gamma_s(t)$ is a weighted linear combination of the parameters v_1, v_2, \dots, v_k ,

$$\gamma_s(t) = c_1(t) v_1 + c_2(t) v_2 + \dots + c_k(t) v_k. \quad (2.12)$$

The functions $c_j(t)$ ($j=1, \dots, k$) are linear functions of t , $c_j(t) = \frac{d}{dt} l_j(t)$ (for further details see Appendix 2).

The spline approximation $\gamma_s(t)$ has an important "minimum" property.

T_k

The quantity $\int_0^{T_k} \gamma_s(t)^2 dt$ can be considered as a measure for the amount of change in the trend of transmission in the entire period $[0, T_k]$ before survey date. As it is the objective to assess changes in the trend of transmission it is important to approximate the unknown transmission function $\lambda(t)$ with a function which changes as little as possible in period $[0, T_k]$. The spline approximation $\lambda_s(t)$ possesses this property. A second reason why application of mathematical splines meets the object of this study is seen here.

Consider the class of continuous functions having a continuous first derivative and possessing the property of preservation of surface. Then $\lambda_s(t)$ belongs to this class and $\lambda_s(t)$ shows a minimum amount of change in transmission in period $[0, T_k]$ before survey date (see Appendix 2, theorem 2.1), thus

$$\int_0^{t_k} \gamma_s(t)^2 dt \text{ is minimal.} \quad (2.13)$$

This property is therefore called *minimum change property* of the trend function $\lambda_s(t)$.

2.1.7 Statistical inference, the trend-change test

In this section a statistical method is proposed for the detection of change in the trend of transmission. The method is based on an analysis of age specific seropositivity rates which are brought forward by a present or past parasitaemia. The cumulative inoculation rate of age group A_j , denoted by p_j , is the proportion of individuals of age A_j who have experienced at least one inoculation in their life.

The detection of change in the trend of the transmission intensity t years before survey date is based on the estimation of the spline approximation $\lambda_s(t)$ which uses estimates \hat{p}_j of the cumulative inoculation rates p_j . The knots of the spline approximation $\lambda_s(t)$ are placed at those time points before survey date which correspond with the mid-points of the age groups A_j . For estimation of the value of the spline approximation $\lambda_s(t)$, t years before survey date, we use the estimator¹⁾

$$\hat{\lambda}_s(t) = l_1(t) \hat{v}_1 + l_2(t) \hat{v}_2 + \dots + l_k(t) \hat{v}_k. \quad (2.14)$$

The statistics $\hat{v}_1, \hat{v}_2, \dots, \hat{v}_k$ are the maximum likelihood (ML) estimators of the parameters v_1, v_2, \dots, v_k ,

$$\hat{v}_j = -\log(1 - \hat{p}_j) \quad (j=1, \dots, k). \quad (2.15)$$

1) A " $\hat{}$ " above a parameter refers to the estimator or estimate of this parameter.

For large sample sizes n_j in age groups A_j ($j=1, \dots, k$) the estimator $\hat{\lambda}_s(t)$ is approximately normally distributed with mean $\lambda_s(t)$ and (asymptotic) variance given by

$$\sum_{j=1}^k l_j(t)^2 \frac{e^{v_j} - 1}{n_j} = \sum_{j=1}^k l_j(t)^2 \frac{p_j}{1-p_j} \frac{1}{n_j}. \quad (2.16)$$

The asymptotic standard deviation of $\hat{\lambda}_s(t)$ is consistently estimated by

$$SD(\hat{\lambda}_s(t)) = \sqrt{\sum_{j=1}^k l_j(t)^2 \frac{\hat{p}_j}{1-\hat{p}_j} \frac{1}{n_j}}.$$

The estimator $\hat{\lambda}_s(t)$ is a weighted linear combination of the statistics v_j ($j=1, \dots, k$); the coefficients $l_j(t)$, $j=1, \dots, k$, are time-dependent functions. The estimators $\hat{\lambda}_s(t_1)$, $\hat{\lambda}_s(t_2)$ of the trend of the transmission intensity in two time points t_1 , t_2 are therefore correlated. In order to compare the values $\hat{\lambda}_s(t_1)$, $\hat{\lambda}_s(t_2)$ an asymptotic statistical test has been used. This test is applied in section 2.2 of this chapter for the detection of differences between the relative extremes of the estimated trend in transmission intensity. Further details with respect to the correlation between $\hat{\lambda}_s(t_1)$ and $\hat{\lambda}_s(t_2)$ and the test of the hypothesis $H_0: \lambda_s(t_1) = \lambda_s(t_2)$ are presented in Appendix 2, section 2.2.2.

To estimate the change in the trend of the transmission intensity t years before survey date the same method of estimation is used,

$$\hat{\gamma}_s(t) = c_1(t) \hat{v}_1 + c_2(t) \hat{v}_2 + \dots + c_k(t) \hat{v}_k. \quad (2.17)$$

The statistic $\hat{\gamma}_s(t)$ is treated as an observed value of a normal random variable having mean $\gamma_s(t)$ and standard deviation,

$$SD(\hat{\gamma}_s(t)) = \sqrt{\sum_{j=1}^k c_j(t)^2 \frac{\hat{p}_j}{1-\hat{p}_j} \frac{1}{n_j}}. \quad (2.18)$$

The estimators $\hat{\gamma}_s(t_1)$, $\hat{\gamma}_s(t_2)$ of the rate of change in the trend of the transmission intensity in two time points t_1 , t_2 are also correlated (for further details see Appendix 2, section 2.2.2).

Trend-change test for a homogeneous population

The rate of change in $\hat{\lambda}_s(t)$, which is a function of the cumulative inoculation rates \hat{p}_j , is used to test the following hypothesis:

H_0 : there is no change in the trend of transmission t years before survey date, $\gamma_s(t) = 0$, against the two sided alternative

H_1 : there is change in the trend of transmission t years before survey date, $\gamma_s(t) \neq 0$.

Using the large sample normal approximation of the distribution of

$$T(t) = \frac{\hat{\gamma}_s(t)}{SD(\hat{\gamma}_s(t))} , \quad (2.19)$$

a two sided p-value can be calculated for each time point t before survey date (with steps of 0.1 year). The resulting "continuous" p-value function is plotted for the detection of periods in which the trend of transmission has changed ($\gamma_s(t) \neq 0$) describing at the same time its level of significance for each time point t before survey date. If the two sided p-value function takes on a value below a critical value α , then it is decided that $\lambda_s(t)$ has changed in the corresponding time point before survey date. The value $\alpha = 0.05$ has been chosen and all time points t are calculated for which

$$\text{p-value} \leq 0.05.$$

This set of time points may consist of several disjunct periods. These periods are considered to indicate a downward or an upward trend in the transmission intensity.

Bruce-Chwatt *et al.* (1973) have reported the results of a serological survey carried out in Mauritius in 1972. Table 2.1 presents the total number of people in the Black River District with sera reactive in malaria IFA tests with *P.falciparum*, *P.vivax*, or *P.malariae* antigen. These data which were analysed using the method of Draper, Voller and Carpenter (1972), are very suitable for the illustration regarding the application of the time-dependent transmission model. The trend of malaria transmission in the model presented here is described by a spline approximation $\lambda_s(t)$ of the transmission function $\lambda(t)$.

Table 2.1 Total number of people with sera reactive in malaria IFA tests with *P.falciparum*, *P.vivax*, or *P.malariae* antigen, Black River District, Mauritius, 1972. Data from 1)

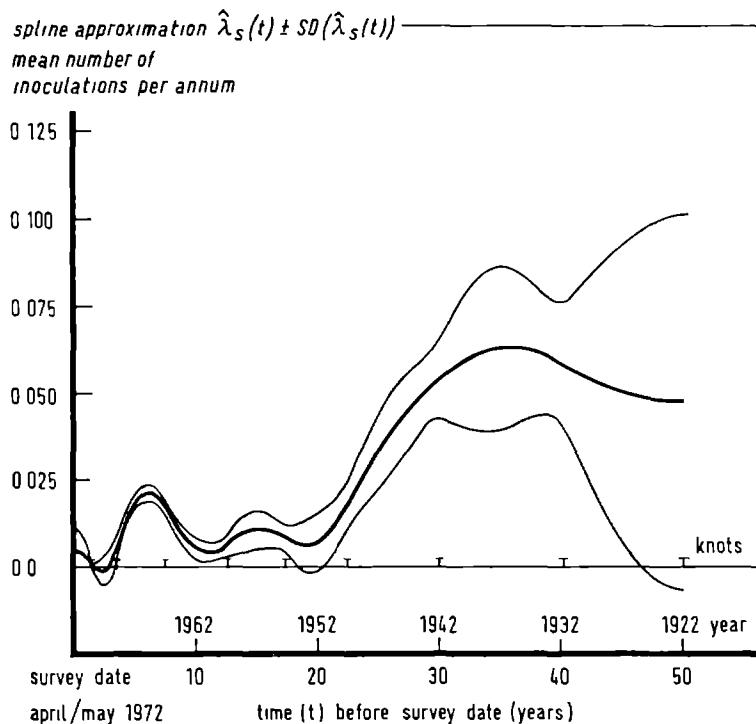
Age group	No-examined	No-positive	%-positive
12-23 mo.	169	1	0.6
2-4 yr.	741	5	0.7
5-9 yr.	2494	183	7.3
10-14 yr.	1164	123	10.6 ²⁾
15-19 yr.	479	70	14.6 ²⁾
20-24 yr.	304	56	18.4
25-34 yr.	167	65	38.9
35-44 yr.	69	46	66.7
45-54 yr.	30	24	80.0

1) Bruce-Chwatt, L.J. *et al* (1973). Sero-epidemiological evidence of eradication of malaria from Mauritius. *The lancet*, 8:547-551. These data are used to illustrate the application of the model.

2) The percentages recorded in the article are 10.5 and 14.7 respectively.

Using the data of table 2.1, fig. 2.4 shows the estimate $\hat{\lambda}_s(t)$ of the trend of the transmission intensity $\lambda_s(t)$, with its standard deviation. The time t before survey date (April/May 1972) has been plotted along the horizontal axis; the mean number of inoculations per annum (the value of $\hat{\lambda}_s(t)$) along the vertical axis. The knots of the spline-trend function $\hat{\lambda}_s(t)$ are placed at the mid-points of the age-groups of table 2.1 (1.5, 3.5, 7.5, 12.5, 17.5, 22.5, 30, 40 and 50 years before survey date). Between two adjacent knots the transmission function $\hat{\lambda}_s(t)$ is a polynomial of degree 2. The knots cannot be read from the curve in fig. 2.4 because in the knots the polynomials fulfil

Figure 2.4 Estimation of the trend of malaria transmission ($\lambda_s(t)$) in Mauritius with its standard deviation. Re-analysis of serological data from Mauritius, Black River District, 1972.



continuity conditions for the polynomials itself and the derivatives. In the period 40-50 years before survey the standard deviation of $\hat{\lambda}_s(t)$ is relatively very large.

Table 2.2 presents the (local) extreme values of the trend function $\hat{\lambda}_s(t)$ and the estimated correlation coefficients of the corresponding estimators. The estimated correlation coefficient between the estimators of the trend of the transmission intensity in the time points 6.0 and 35.8 years before survey date ($\hat{\lambda}_s(6.0)$ and $\hat{\lambda}_s(35.8)$ are local extreme values) is 0.00.

Table 2.2 Local extreme values of the trend of the transmission intensity $\hat{\lambda}_s(t)$ with its standard deviation and correlation coefficients between these extreme values¹⁾ (Mauritius, Black River District, 1972).

Transmission function $\hat{\lambda}_s(t)$		Correlation coefficients between extreme values							
Time before survey date (years)	Extreme value \pm SD	Time before survey date (years)							
		0.0	2.2	6.0	11.0	15.2	18.8	35.8	50.0
0.0	0.006 \pm 0.006		-0.90	0.48	-0.15	0.02	0.00	0.00	0.00
2.2	-0.002 \pm 0.003	-0.90		-0.69	0.22	-0.04	0.00	0.00	0.00
6.0	0.021 \pm 0.002	0.48	-0.69		-0.55	0.04	0.01	0.00	0.00
11.0	0.004 \pm 0.003	-0.15	0.22	-0.55		-0.27	-0.17	-0.02	0.01
15.2	0.010 \pm 0.006	0.02	-0.04	0.04	-0.27		-0.44	-0.04	0.02
18.8	0.006 \pm 0.007	0.00	0.00	0.01	-0.17	-0.44		0.25	-0.12
35.8	0.063 \pm 0.023	0.00	0.00	0.00	-0.02	-0.04	0.25		-0.54
50.0	0.048 \pm 0.054	0.00	0.00	0.00	0.01	0.02	-0.12	-0.54	

1) Unit: mean number of inoculations per annum

Table 2.3 shows the results of the pair-wise comparison between the extreme values of the transmission function $\hat{\lambda}_s(t)$. A difference between any two extreme values of $\hat{\lambda}_s(t)$ is treated as an observed value of a normal random variable with known standard deviation. The corresponding "p-values" are presented in table 2.3. The p-values are used here for exploratory purposes only i.e. to examine observed extreme values of $\hat{\lambda}_s(t)$. A clear pattern is seen; differences between any two extreme values appear in relation with the extreme values 0.021 and 0.063 at the time points 6.0 and 35.8 years respectively before survey date. These extreme values are different from all other extreme values with one exception i.e. the extreme value (0.048) 50 years before survey date.

Table 2.3 Pair-wise comparison of all extreme values of the trend of the transmission intensity $\hat{\lambda}_s(t)$, (Mauritius, Black River District, 1972).

Time before survey date (years)	<i>p</i> -values ¹⁾ used to detect differences between extreme values							
	Time before survey-date (years)							
	0.0	2.2	6.0	11.0	15.2	18.8	35.8	50.0
0.0	NS	**	NS	NS	NS	*	NS	
2.2	NS	****	NS	NS	NS	**	NS	
6.0	**	****	***	(*)	*	(*)	NS	
11.0	NS	NS	***	NS	NS	*	NS	
15.2	NS	NS	(*)	NS	NS	*	NS	
18.8	NS	NS	*	NS	NS	*	NS	
35.8	*	**	(*)	*	*	*	NS	
50.0	NS	NS	NS	NS	NS	NS	NS	

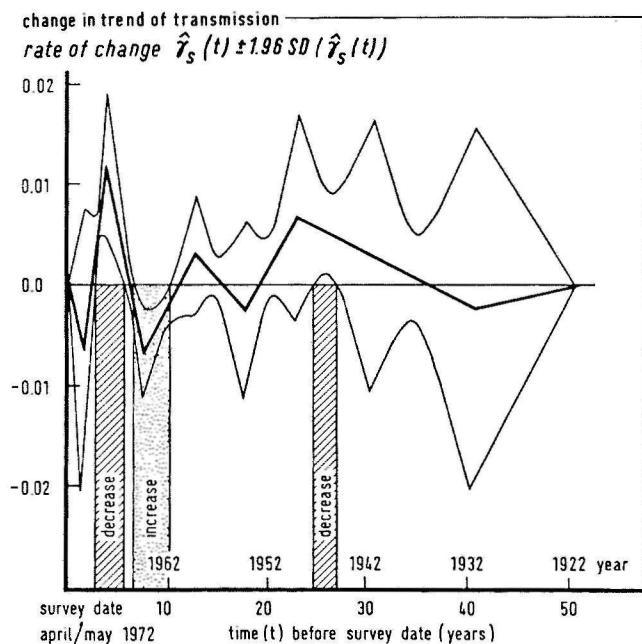
1) A p-value is used here in an exploratory way.

Meaning of symbols:

NS	(not significant)	0.10 < p
(*)	(indication)	0.05 < p \leq 0.10
*	(significant)	0.01 < p \leq 0.05
**	"	0.001 < p \leq 0.01
***	"	0.0001 < p \leq 0.001
****	"	p \leq 0.0001

Fig. 2.5 shows for each time point t before survey date the estimate $\hat{\gamma}_s(t)$ of the rate of change in the trend of transmission, that is the rate of change of $\hat{\lambda}_s(t)$. The graph of $\hat{\gamma}_s(t)$ is a broken line, its pieces join in the knots. The direction of the conventional time scale has been reversed. So a positive and a negative value of $\hat{\gamma}_s(t)$ corresponds with a decrease and an increase respectively of the estimate of the trend in transmission (see fig. 2.4 and fig. 2.5). Three periods are found with time points t for which the rate of change $\hat{\gamma}_s(t)$ is statistically different from zero. As prescribed by the model the rate of change in $\hat{\lambda}_s(t)$ is zero in the endpoints of the curve ($\hat{\gamma}_s(0) = \hat{\gamma}_s(50) = 0$, $SD(\hat{\gamma}_s(0)) = SD(\hat{\gamma}_s(50)) = 0$).

Fig. 2.5 Estimation of the rate of change ($\hat{\gamma}_s(t)$) in the trend of the transmission intensity with approximate 95% confidence limits. Three intervals are found with a statistically significant change. Re-analysis of serological data from Mauritius, Black River District, 1972.



A more detailed picture of the change in the trend of transmission is obtained by presenting the estimate $\hat{\lambda}_s(t)$ and the p-value function associated with the detection of change in $\lambda_s(t)$ in one fig. (fig. 2.6). This type of presentation will be used as a standard method for the detection of change in the trend of the transmission intensity in the other applications (see chapter 5). In interpreting fig. 2.6 three successive steps have to be made:

First: the curve in the upper-half of the figure represents the estimate $\hat{\lambda}_s(t)$ of the spline approximation of the transmission intensity function.

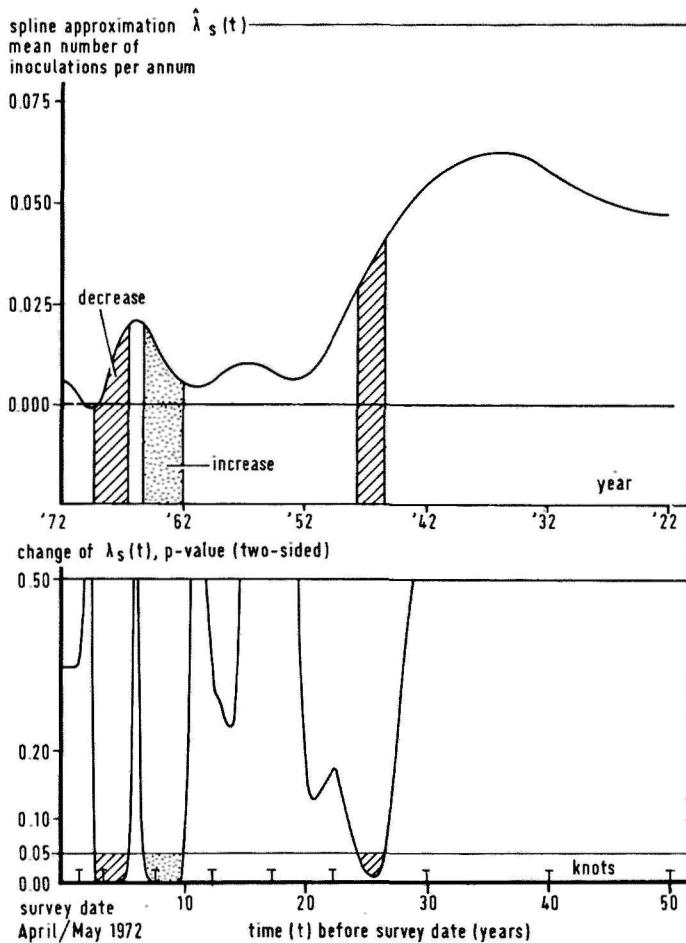
Second the curve in the lower-half of the figure presents the two-sided p-value function for the detection of periods of change in $\lambda_s(t)$.

Third: the shaded areas in the upper-half of the figure indicate those periods in which the p-value function takes on a value below the critical level 0.05.

A statistically significant decrease in the trend of the transmission intensity $\lambda_s(t)$ is found in the period 2.6-5.4 years and 24.3-26.5 years before survey date; an increase in the trend is indicated in period 6.6-10.0 years before survey date.

Table 2.4 (see page 26) shows estimated correlation coefficients between estimates $\hat{\gamma}_s(t)$ with time points t chosen in these intervals. The time points 2.9, 9.0, 25.5 years before survey date correspond to estimates $\hat{\gamma}_s(t)$ with the highest statistically significant change in that interval. The correlation coefficient between the estimates of the change in transmission 2.9 and 9.0 years before survey date is -0.81. In general an over estimation of the increase 9.0 years before survey date involves an over estimation of the decrease 2.9 years before survey date. However, table 2.4 shows that there are also positive correlations between the changes $\hat{\gamma}_s(t)$ in these intervals. This implies that there exist time points t_1 in interval 2.6-5.4 years before

Figure 2.6 Standard representation¹⁾ for the detection of change in the trend of transmission; an illustration using data from Mauritius.



¹⁾ legenda, $\lambda_s(t)$ = spline-trend of the transmission intensity

survey date and t_2 in interval 6.6-10.0 years before survey date with a statistically significant decrease and increase in trend of transmission respectively which are uncorrelated. The change in transmission 25.5 years before survey date $\hat{\gamma}_s$ (25.5) shows only a very weak correlation with the change 2.9 and 9.0 years respectively before survey date (correlation coefficient 0.11 and 0.09 resp.).

Table 2.4 Correlation coefficients between estimates $\hat{\gamma}_s(t)$ of changes in the trend of the transmission intensity at several time points before survey date¹⁾ (Mauritius, Black River District, 1972).

Time before survey date (years)	Time before survey date (years)								
	First interval			Second interval			Third interval		
	2.6	2.9	5.4	6.6	9.0	10.0	24.3	25.5	26.5
2.6		0.35	-0.82	-0.39	0.11	0.21	0.05	0.05	0.02
2.9	0.35		0.13	-0.74	-0.81	-0.19	0.12	0.11	0.05
5.4	-0.82	0.13		0.27	-0.47	-0.53	-0.11	-0.10	-0.04
6.6	-0.39	-0.74	0.27		0.60	-0.31	-0.30	-0.27	-0.12
9.0	0.11	-0.81	-0.47	0.60		0.51	0.10	0.09	0.03
10.0	0.21	-0.19	-0.53	-0.31	0.51		0.57	0.51	0.22
24.3	0.05	0.12	-0.11	-0.30	0.10	0.57		0.78	0.21
25.5	0.05	0.11	-0.10	-0.27	0.09	0.51	0.78		0.77
26.5	0.02	0.05	-0.04	-0.12	0.03	0.22	0.21	0.77	

1) The periods [2.6,5.4], [6.6,10.0] and [24.3,26.5] denote 3 intervals with a statistically significant change $\hat{\gamma}_s(t)$ in transmission. The time points 2.9, 9.0, 25.5 years before survey date correspond to estimates $\hat{\gamma}_s(t)$ with the highest statistically significant change in the corresponding interval.

2.3 CORRECTION FOR AGE SPECIFIC MALARIA DEATH RATE AND OTHER CAUSES OF DEATH

A cross-sectional survey will always miss those individuals who have died

from malaria or other causes of death before the day of the survey. Thus the proportion of the individuals of age A_j alive at the day of the survey who have experienced at least one infection with malaria - i.e. the cumulative inoculation rate \hat{p}_j in a cross-sectional survey - is a biased estimate of the proportion of the individuals born A_j years before survey date who have had malaria or would have contracted a malaria infection if they had survived until the day of the survey and had been exposed to the same inoculation risk as those who stayed alive. If reliable figures are available on the malaria death rate of the various age groups and if we may assume 'equal inoculation risk' for individuals who have died from other causes of death and individuals who survived until survey date (equal inoculation risk to be specified in detail below), then the value \hat{p}_j might be corrected without knowing the non-specific malaria death rate.

At survey date the following eventualities for an individual born A_j years before survey date must be considered:

D^m	=	died from malaria before survey date
D^o	=	died from other cause before survey date
L	=	is still alive at survey date
M	=	had malaria, or would have had malaria if had survived until survey date and had been exposed to the same inoculation risk as the survivors.

let $p_j[x] =$ proportion of the individuals born A_j years before survey date who experienced eventuality x

The specific death rates and survival rate are defined as follows¹⁾:

d_j^m	=	$p_j[D^m]$	=	age specific malaria death rate
d_j^o	=	$p_j[D^o]$	=	age specific non-malaria death rate
l_j	=	$p_j[L]$	=	age specific survival rate.

Furthermore three "cumulative inoculation rates" can be distinguished:

¹⁾ Note that these rates are cumulative ones

$$\begin{aligned}
 p_j &= p_j[M|L] = \text{cumulative inoculation rate in the survivors} \\
 p_j^o &= p_j[M|D^o] = \text{"cumulative inoculation rate in } D^o \text{ group"} \\
 p_j^c &= p_j[M] = \text{corrected cumulative inoculation rate}
 \end{aligned}$$

The cross-sectional survey provides an estimate \hat{p}_j of p_j . The objective here is to correct \hat{p}_j for the death rates d_j^m , d_j^o and to derive an estimate of p_j^c .

The corrected inoculation rate $p_j[M]$ is equal to

$$p_j[M] = p_j[M|L]p_j[L] + p_j[M|D^m]p_j[D^m] + p_j[M|D^o]p_j[D^o].$$

As $p_j[M|D^m] = 1$ we have

$$p_j^c = p_j[1_j + d_j^m + p_j^o d_j^o].$$

From the definition of 1_j , d_j^m and d_j^o follows

$$1_j + d_j^m + d_j^o = 1.$$

Substitution of $1_j = 1 - d_j^m - d_j^o$ in the formula for p_j^c results in the general formula

$$p_j^c = p_j + d_j^m(1-p_j) + d_j^o(p_j^o - p_j). \quad (2.20)$$

If equal inoculation risk may be assumed for individuals who have died before survey date however not from malaria and individuals who have survived until survey date then the following relation holds

$$p_j^o = p_j. \quad (2.21)$$

It follows at once (see (2.20)) that the corrected inoculation p_j^c in this situation is equal to

$$p_j^c = p_j + d_j^m(1-p_j). \quad (2.22)$$

Consequently in order to account for the toll of death due to malaria, one should use the corrected value \hat{p}_j^c instead of \hat{p}_j in performing the analysis,

$$\hat{p}_j^c = \hat{p}_j + d_j^m(1-\hat{p}_j). \quad (2.23)$$

Actually p_j^o might be larger than p_j because malaria can be an indirect cause of death. Therefore when only correction for specific malaria death rate is applied the 'true' corrected inoculation rate p_j^c is still probably underestimated. If those individuals who died from indirect causes of malaria can be included in the age specific death rate d_j^m for malaria, then the corrected value \hat{p}_j^c should be based on this figure.

2.4 DISCUSSION

2.4.1 General remarks regarding methodology

The time-dependent transmission model presented in this chapter concerns the assessment of the trend of malaria transmission by the use of cross-sectional data. In principle, however, the model can be applied to any field of research which directly or indirectly involves numerical and/or statistical inference of the parameter $\lambda(t)$ of a catalytic model. This can be substantiated as follows.

A population is considered in which the uninfected individuals are conceived as subjected to a force of infection which changes them to infected individuals. Let $p(t)$ be the fraction of individuals infected at time t . Assuming a time-dependent force of infection acting on the uninfected individuals and no recovery of those already infected, the catalytic differential equation can be written as (*Muench (1949)*)

$$\frac{dp}{dt} = \lambda(t)(1-p(t)) \quad 0 \leq p(t) < 1,$$

with $\lambda(t)$ as the time-dependent catalytic force.

Such an equation for $\lambda(t)$ does not imply that a non-stationary Poisson process is being dealt with. Hence $\lambda(t)$ does not have to represent the mean number of

events per annum inflicted on one individual of the population at time t . Nevertheless the parameter $\lambda(t)$ can still be estimated by a procedure analogous to the procedure described in the time-dependent transmission model. It is readily verified that the catalytic equation can be rendered as

$$\lambda(t) = \frac{d}{dt} [-e^{\lambda(t)} \log(1-p(t))].$$

Hence the first derivative of a natural cubic spline approximation of the function $-e^{\lambda(t)} \log(1-p(t))$ with knots in the data points $(t_j, -e^{\lambda(t_j)} \log(1-p(t_j)))$ ($j=0, 1, \dots, k$) can be used for the assessment of the catalytic force $\lambda(t)$. It is exactly this type of inference that has been used in the time-dependent transmission model. Thus the methodology proposed in this chapter may be used to study the trend of all those diseases for which the catalytic model is assumed to be valid (especially cohort-studies). The parameter $\lambda(t)$ can be interpreted as the mean number of effective contact, per annum inflicted on one individual of the population if the assumptions required for a non-stationary Poisson process and a homogeneous population are presupposed (see section 2.1.2). Effective contact has the meaning used by Muench and Wade Frost (Muench, 1959):

"a contact sufficient to produce infection if the subject is susceptible".

Examples of other diseases than malaria for which the catalytic model has been used for the analysis of cross-sectional serological data are yellow-fever (Muench (1959)). $\lambda(t)$ is independent of time, $\lambda(t) = 1$, tuberculosis (Lafaye (1976)): $\lambda(t)$ is exponential, $\lambda(t) = e^{c+st}$) and hepatitis A (Schenzle, Dietz and Frosner (1979)): $\lambda(t)$ is asymmetric logistic function, $\lambda(t) = c - c/(1 + \exp \alpha(t+0)))$.

The choice of $\lambda(t)$ in these three examples is purely heuristic. If the data are not inconsistent with the model its choice is justified. It is obvious that these 3 models can not be applied in general for the assessment of a

fluctuating trend in the frequency of effective contacts. Generally these 3 models exclude each other. However the model introduced here " $\lambda(t) = \lambda_s(t)$ = spline approximation of $\lambda(t)$ " can be applied to any of these diseases. It is applicable irrespective of the true form of the time-dependent force of infection $\lambda(t)$. If in a homogeneous population it is assumed that evidence of exposure to infection is definite during life and mortality is negligible, then the time-dependent transmission model can be used in principle for the analysis of cross-sectional serological data of any disease.

Although the methodology is neither restricted to the study of the trend in the intensity of malaria transmission nor to the analysis of only cross-sectional data (in fact it may be better used for the determination of trends in other diseases, especially cohort-studies) the remainder of this discussion is devoted to malaria. The applicability of the model to other fields of interest and the analysis of other data will be further discussed in chapter 6.

2.4.2 The concept of inoculation

Macdonald has established the inoculation rate (h) as a numerical measure of the intensity of transmission. However, some misunderstanding may arise about the meaning of the word inoculation. Macdonald (1957) defines the inoculation rate " h " as the mean daily number of bites inflicted on one individual by mosquitoes infected with sporozoites which are actually infective. The term "actually infective" is very interesting. Macdonald illustrates the discrepancy between what he calls "total" and "successful" inoculations by comparing calculations based on entomological data (i.e. the man-biting rate "m.a." and the sporozoite rate "s") with estimates derived from an age-specific breakdown of infant parasite rates. Since infants are presumably non-immune, Macdonald thus appears to equate successful inoculations with inoculations

capable of causing parasitaemias in non-immune subjects.

Swaroop (1966) , on the other hand, points out that the term "actually infective" should be interpreted as actually succeeding in establishing infection which may depend on the immune response of the average individual. Although he repeats Macdonald's example of the infant parasite rate to illustrate this point, he goes on to state that the discrepancy noted may be greater in areas where immunity is high. "Evidence does not extend, however, to proving whether this is due to restriction of gametocyte output, and hence low sporozoite infections of mosquitos, or to true resistance by the recipient of the sporozoites" (quoted from Swaroop (1966), op.cit. chapter 12, p. 132). Swaroop gratefully acknowledge Macdonald for his kindness in reading chapter 12 of his book. Nevertheless Swaroop's concept of the inoculation phenomenon might be different from that of Macdonald.

The concept of inoculation in this study may differ from those quoted above. Normally inoculation is defined as the receipt of a certain dosage of an infectious agent. Under natural conditions an inoculation with sporozoites is an event which is not directly evident. It is only in an indirect way that such an inoculation may become apparent, i.e. not by the multiplication of parasites in the liver but by the later occurrence of a parasitaemia. For this reason and in order to avoid possible interference due to immunity acquired from earlier infections with malaria parasites (including sporozoites) in this study an *inoculation* is defined as an event leading to a parasitaemia if it occurs on a previously uninfected individual. Consequently such an inoculation can also occur in a previously infected individual. In that case, however, such an event does not necessarily lead to a fresh parasitaemia; in this situation the occurrence of a fresh parasitaemia will depend on the absence of immunity in the recipient. In the model the concept of inoculation as such is independent from immunity factors.

Although in the definition of the event of inoculation "previously uninfected" is preferred rather than "actually infective" or "non-immune", Macdonald's conception of inoculation may not be essentially different from that used in this study so far.

In case that the population is provided with prophylactic drugs a difference in interpretation might easily occur. The transmission function $\lambda(t)$, which measures the mean number of inoculations per annum inflicted on the individual, should be capable of determining the effect of antimalarials on the interruption of malaria transmission. Therefore and also since the measurements aim to detect parasitaemias, an inoculation is equated with the receipt of a quantity of sporozoites capable of causing a parasitaemia in a previously non infected individual who may or may not have used antimalarials. Consequently on purpose for this study "an inoculation" is deliberately considered to be dependent of the natural resistance of the recipient against a primary parasitaemia and includes here also the protection by use of antimalarials. This implies that the daily inoculation rate inflicted on one individual is influenced by the provision of anti malarial drugs. It is in this respect that the concept of inoculation as defined here might be different from that of Macdonald.

Muench (1959) uses the term *effective contact*¹⁾ in the general context of a force of infection acting on the population. If we consider the term *susceptible* also as dependent on the protection of the subject by prophylactic drugs, then the concept of inoculation used here does not seem to differ in essence from Muench's conception of effective contact.

¹⁾ Effective contact is a contact sufficient to produce infection if the subject is susceptible.

2.4.3 The transmission assumptions

The four transmission assumptions are essential for the interpretation of $\lambda(t)$. Assumption 1 relates to an ever present positive probability of receiving an inoculation; this is realistic for any infectious disease. This would become zero when eradication has been achieved.

A few comments have to be made which regard to assumption 2. A mosquito may take a blood meal which consists of several bites in a very short time interval on the same host. If the quality of sporozoites of these bites is sufficient to induce a primary parasitaemia, then these bites together are considered as one inoculation. When an inoculation is considered in this way assumption 2 is acceptable.

Assumption 3 is realistic if it is assumed that the probability of receiving an inoculum does not depend on the occurrence of earlier inoculations; for instance the infection with malaria does not interfere with or predispose for a fresh reinoculation. This assumption seems to be acceptable for malaria and many other infectious diseases.

Assumption 4 requires equal risks for all individuals of the population under study living in a certain area. Of course this assumption is only an approximation of the reality; it ignores the individual characteristics of non-specific immunity, the possible presence of maternal antibodies in newborn children, habits of life, migratory habits etc. It implies that the risk of receiving an inoculum is independent of age i.e. it is assumed that there is no age preference with regard to mosquito bites. If the study is restricted to a population living in a relatively small area such as a village, and if migration as a factor is excluded assumption 4 is acceptable for the present purpose.

In a homogeneous population the cumulative inoculation rate p_j increases with age. In this model it is presupposed that the cumulative inoculation rate

is equivalent to the seropositivity rate. This would imply that the seropositivity rate also increases with age. An increase of the seropositivity rate with age is a commonly observed phenomenon in homogeneous populations and is assumed to be the case in the following chapters.

2.4.4. The transmission function, explanatory variables

The transmission function $\lambda(t)$ describes the instantaneous force of potential malaria infections, that is the mean number of inoculations per annum inflicted on an individual t years before survey date. The characteristics of the mosquito and the human population at time t before survey are the two mutually dependent main factors which determine the value of the transmission function t years before survey. An adapted notation derived from the detailed investigation of Macdonald (1957) will be used, this has been widely referred to in the literature on malaria epidemiology. Let m , a , s , b' be defined by:

m : the mosquito density in relation to man;

a : the average number of individuals bitten by one mosquito in one day,
i.e. the man-biting habit;

s : the proportion of mosquitoes with sporozoites in their salivary glands;

b' : the proportion of the mosquitoes with sporozoites in their salivary glands,
which are capable of inducing a parasitaemia in a previously uninfected
human.

So if m , a , s , en b' are considered as functions of time, $\lambda(t)$ is approximately equal to

$$\lambda(t) = 365 m(t) a(t) b'(t) s(t),$$

where t denotes a time point t years before survey date.

Macdonald's formula for the inoculation rate is "h=mabs" with b as the proportion of mosquitoes having sporozoites in their salivary glands that are actually infectious. It follows that the relation between the transmission function $\lambda(t)$ and the inoculation rate h of Macdonald if considered a function of time is approximately given by

$$\lambda(t) = 365 \frac{b'(t)}{b(t)} h(t).$$

Generally the man biting habit $a(t)$ is assumed not to change in time. The explanatory variables $m(t)$, $s(t)$ and $b'(t)$ may change substantially due to various factors like changes in temperature and climate, ecological changes and successful eradication programmes. The variable $b'(t)$ depends also on the resistance against a primary parasitaemia present in the population at time t before survey date. This resistance may change in time due to changes in health condition of the population and the use of anti malarial drugs. It should be noticed that, although $b'(t)$ does not depend on the specific immunity of the recipient at time t it may be influenced by the immunity of the population at time $t+\Delta t$. This might result in a low density of fertile gamocytes and hence in scant sporozoite infections of mosquitoes at time t.

2.4.5 Spline approximations

The spline function $\lambda_s(t)$ is an approximation of the transmission function $\lambda(t)$. It reflects certain aspects of the trend of the transmission intensity. The approximation can be made more accurate by choosing the knots more closely together. However, the kind of data and the sample size will determine to a very large extent the place of the knots. There is a kind of analogy in drawing a histogram representing a frequency distribution and choosing a spline approximation $\lambda_s(t)$ of the transmission function $\lambda(t)$. The width and choice of the class intervals of a histogram determines to a large extent the

kind of picture that is obtained from the unknown frequency distribution. The choice of the knots determines in an analogous way the kind of picture of the trend of the unknown transmission function. Seasonal variation is only reflected in the spline approximation $\lambda_s(t)$ if the distance between the knots is a few months. If the distance between the knots is one, two, three or even more years only certain aspects of the long term course of the transmission function $\lambda(t)$ are described by $\lambda_s(t)$. When the distance between the knots is one year, the spline approximation $\lambda_s(t)$ describes the annual trend of the transmission intensity.

When the method of analysis is used for exploratory purposes, then it is recommended to perform the analysis several times with the knots placed at different points on the time axis. Each curve of a spline approximation $\lambda_s(t)$ of the transmission function $\lambda(t)$ will show another aspect of the trend of the transmission intensity. If there is some apriori knowledge on the time points before survey date where the transmission may have changed substantially then it is recommended to place knots at these time points. In serological surveys, periods longer than one year have often to be used as the distance between two knots. In that case there is an argument to use an integer number of years as distance between the knots in order to eliminate the influence of the seasonal variation on the trend of the transmission. If the distances between the knots are larger than one year and variable, great care is needed in the interpretation of $\lambda_s(t)$. Long term spline-trends based on unequal periods between the knots can be misleading, since then different type of trends are being compared.

It should be noted that negative values of the spline approximation $\lambda_s(t)$ may occur. Therefore negative values of $\lambda_s(t)$ are seen in the graphical representations. However, the integral of $\lambda_s(t)$ between the knots represents exactly the mean number of inoculations in this period, i.e. the area between the curve

and the horizontal axis is taken as negative for intervals where $\lambda_s(t)$ is negative. To assess the trend of the transmission intensity, the property of preservation of surface between the knots is a favourable property of the spline approximation $\lambda_s(t)$. However, there might be some less favourable side effects as well. For example, intervals adjacent to periods in which $\lambda_s(t)$ is negative will show artificial high levels of $\lambda_s(t)$ due to that property of preservation of surface.

The function $\gamma_s(t)$ describes the rate of change in the spline trend $\lambda_s(t)$. This function $\gamma_s(t)$ consists of piecewise defined polynomials of degree 1 which join in the knots. By definition $\gamma_s(0) = \gamma_s(T_k) = 0$ and accordingly $SD(\hat{\gamma}_s(0)) = SD(\hat{\gamma}_s(T_k)) = 0$. At the knots T_1, T_2, \dots, T_{k-1} the standard deviations are larger than in between the knots (see fig. 2.5). This phenomenon should be compared with that of regression lines, where the estimation of the value of the regression line occurs with greater accuracy in the middle of the regression line than at the end points.

The total amount of the rate of change in the spline approximation $\lambda_s(t)$ in the period under consideration is minimal in comparison with all other possible functions (having a continuous first derivative) which might be used to describe the trend of the transmission intensity and which possess the property of preservation of surface between the knots. The main purpose of this study is the detection of change in the trend of transmission. Therefore from a methodological point of view this "minimum change property" of $\lambda_s(t)$ is a strong optimal property, it particularly explains why the spline approximation $\lambda_s(t)$ has been chosen to assess the trend of the transmission intensity.

2.4.6 The p-value function

In the model, this part takes into account the statistical fluctuations

in the fraction of individuals who have experienced at least one inoculation and who belong to a specified age group A_j ($j=1, \dots, k$). This fraction was called "the cumulative inoculation rate (\hat{p}_j)". Even the examination of an entire small community is still considered to involve a sample only. This is so in the sense that the fraction actually found positive should be considered as an estimate of the expected fraction positive. The latter is based on the existing risk of infection as measured by the transmission intensity $\lambda(t)$.

The p-value function is used for the *detection* of periods of change in the trend of the transmission intensity as well as for the description of the significance level of the test result. The choice of the value α is of course arbitrary. If a significance level $\alpha = 0.05$ is used, as has been done, then testing the hypothesis that no change in the trend of transmission has occurred in a preassigned time point t years before survey date would give us misleading results in approximately five out of every 100 occasions in case the hypothesis is true. In practical applications, however, a change in trend of transmission is not only tested for a fixed preassigned time point t years before survey date. Often it is the intention to survey the situation at each individual time point. Then the probability of erroneously rejecting the null hypothesis "no change in trend of transmission at any time t " will be higher than 5%. This error rate will generally increase with the number of age-groups included in the analysis. Therefore if we use a large number of age groups, say larger than 10, then it should be considered to choose a smaller value of α . At a later stage of the study we intend to substitute the time point related statistical inference regarding a change in trend of transmission by a simultaneous statistical procedure. Furthermore the methodology will be adapted to analyse individual recorded data instead of grouped data.

2.4.7 *Comments on the application of the model*

A re-analysis of sero-epidemiological data from Mauritius is performed in

order to illustrate the model's application and to evaluate the results against the well documented history of the epidemiological course of malaria transmission in Mauritius. Before discussing the results the main lines of the epidemiological course of transmission reported by *Bruce-Chwatt et al. (1973)* is given below.

Sir Ronald Ross visited Mauritius in 1907 and recommended the main lines for the control of malaria. During the next 40 years these methods, used mainly in urban areas, contributed to a considerable decrease of the disease, which was confined largely to the rural coastal belt, although small outbreaks of great intensity occurred periodically in the higher areas. In 1949 a malaria-eradication pilot scheme was set up. The results of the anti-malaria operations showed a fall in the number of cases of malaria from 46,000 in 1948 to 6,000 in 1950, a corresponding decrease of the spleen rate from 35% to 2.5% and of the parasite rate from 9.5% to 0.1%. There was also a dramatic decrease of the crude death-rate and of the infant mortality rate. During the nineteen fifties small outbreaks of malaria continued.

The total coverage of DDT insecticide spraying ceased in 1958, but intensive malaria surveillance introduced in 1960 revealed that cases of malaria were more common than previously estimated. However, the relatively large number of cases (1179) found in 1960 decreased to 955 in 1961 and to 226 in 1962. Most of these were found in the districts of Black River and Grand Port, and it was confirmed that transmission of malaria continued on a small scale in these parts of the island. Residual DDT spraying of malarious foci was reinstituted in these areas, and improved surveillance measures resulted in the further fall of the number of cases of malaria to 30 in 1963, 20 in 1964, 14 in 1965 and 12 in 1966.

In 1966 the spraying operations were limited to occasional spraying, but surveillance activity continued unremittingly. During the period 1963-71 only

11 of 148 cases of malaria were definitely due to local transmission. The last such possible case observed in 1968 when the whole island entered into the maintenance phase of malaria eradication, was later classified as cryptic. According to the criteria of the World Health Organization one of the requirements for the achievement of malaria eradication is the absence of indigenous cases of malaria for three years providing that there is an adequate surveillance system. Of the adequacy of the local malaria surveillance system there can be no doubt, and measures have prevented any reintroduction of malaria from outside the island. Thus Mauritius became one of the few countries in Africa where malaria eradication was successful.

The spline approximation of the transmission function $\hat{\lambda}_s(t)$ of fig. 2.4 reflects the main lines of the epidemiological course of malaria on Mauritius as reported by *Bruce-Chwatt et al. (1973)*. The transmission function $\hat{\lambda}_s(t)$ shows a statistically significant decrease in period 24.3 - 26.5 years before survey date that is roughly in the years 1946 - 1947 (see fig. 2.6). The actual drop occurred in the period 1949 - 1950, which is close to the period indicated by the analysis. It should be noted that the distances between the knots are unequal and very large 17.5 - 22.5 and 22.5 - 30 years respectively before survey. This means that $\lambda_s(t)$ describes only the long term trend in transmission intensity in the period 17.5 - 30 years before the survey. Therefore it cannot be required that in this application the methodology should exactly indicate the short term decrease in the period 1949 - 1950. Furthermore the analysis is based on the mid-points of the age groups which of course is a potential source of error, especially in areas with primitive registration services.

The rate of change in the trend of the transmission intensity $\gamma_s(t)$ is described by straight lines which join in the knots. The graphical presentation of the standard deviations of the estimates $\hat{\gamma}_s(t)$ shows a remarkable course

(see fig. 2.5). At the knots the values of the standard deviations are larger than in between the knots. As has already been pointed out, this phenomenon is inherent to the estimation of values of a regression line. Another point of interest is the value of the rate of change $\hat{\gamma}_s(t)$ at the end points of the period under investigation. By definition the rate of change $\hat{\gamma}_s(t)$ is zero at the end points $t = 0$ and $t = T_k$. It is for this reason and the linearity of $\hat{\gamma}_s(t)$ as a function of time that the p-value function takes on a constant value in the first and the last interval defined by the knots respectively (see fig. 2.6; in period 0 - 3.5 yr before survey date the p-value function does not depend on t).

In analysing the Mauritius data the age specific malaria death rates have not been taken into account. The malaria death rate had decreased after 1949. If reliable figures are available on the malaria death rates of the various age groups, it is possible to correct the estimate \hat{p}_j of the cumulative inoculation rate of the age group A_j and to account for the toll of death in performing the analysis (see section 2.3). Disregarding the age specific malaria death rates may cause biased results. It may have a negative effect on the power of the method to detect a downward trend of the transmission intensity. On the other hand it might also induce an artificial decrease in the spline approximation $\lambda_s(t)$.

The spline approximation $\lambda_s(t)$ shows a significant increase and a decrease in period 6.6 - 10.0 years and in period 2.6 - 5.4 years respectively before the survey. The decrease seems not contradictory to the malaria history in Mauritius, however the temporary peak in transmission around the year 1966 does not agree with the course of transmission as reported by *Bruce-Chwatt et al.* (1973). The only indication that a small raise in transmission might have occurred is the remark that in 1966 the spraying operations were limited to occasional spraying. However, no evidence is available for a rise

in transmission in the previous years. How can one explain the temporary increase in the spline-trend function $\lambda_s(t)$ in the years 1962/1965? Relations between the estimates cannot account for the temporary peak about 6 years before the survey. The transmission assumptions seem to be acceptable. However, in the application of the model it has been assumed that a method is available to distinguish between those individuals who experienced at least one inoculation and those who did not receive an inoculum. Due to the normally occurring fading of antibody production following antigenic stimulation, the IFA test for malaria is not capable to distinguish adequately between these two alternatives. This could offer an explanation of the temporary increase in the spline-trend function $\hat{\lambda}_s(t)$. The fading of antibodies after an infection is taken into account in chapter 4. There the unexplained increase in the trend of the transmission intensity in the years before 1966 will be corrected.

A further limitation of the model presented in this chapter was the assumption that a homogeneous population was under study. As a first approximation the assumption of equal risk of receiving an inoculum for all individuals is acceptable for small villages. However, for large populations like Mauritius this seems unrealistic. The next chapter deals with the problem of the determination of the trend in transmission intensity in a non-homogeneous population.

2.5 SUMMARY

A stochastic model is introduced for the determination of the trend of malaria transmission in a homogeneous population. The model takes into account the time-dependent risk of receiving an inoculation and is based on the analysis of estimates of grouped cumulative inoculation rates obtained from one cross-sectional survey. Based on four transmission assumptions and the

inoculation concept a transmission function $\lambda(t)$ is defined which describes the mean number of inoculations per annum inflicted on one individual t years before survey date. Mathematically $\lambda(t)$ presents the intensity function of a non-stationary Poisson process. A spline approximation $\lambda_s(t)$ of the transmission function $\lambda(t)$ is used as a measure of the trend in transmission intensity. Two properties of $\lambda_s(t)$ i.e. the *property of preservation of surface* and the *minimum change property* ensure that $\lambda_s(t)$ is very suitable to describe all kinds of changes in the trend of the transmission intensity. A statistical test is presented for the detection of change in the trend of transmission ("trend-change test"). The model presented here provides a *general parametrisation* for the catalytic model with a time-dependent force of infection.

The model can be applied to reconstruct the epidemiological course of any infectious disease for which the transmission assumptions are sufficiently realistic and for which the survey data provide the means to distinguish between those individuals who have experienced an infection from those who have not. If this information is obtained from serological examination then it has to be assumed that "being positive in a serological test" is equivalent to "having experienced at least one infection".

Finally, a re-analysis is presented of the sero-epidemiological data from Mauritius as an illustration of the model's application. The results are evaluated against the well documented history of the epidemiological course of malaria transmission on Mauritius. The estimated spline approximation of the transmission function reflects fairly satisfactorily the main line of the epidemiological course of malaria as reported by Bruce-Chwatt. However, no evidence is available for the temporary increase in transmission in the years 1962/1965 which was found as a result of the analysis.

THE TREND OF TRANSMISSION IN A NON-HOMOGENEOUS POPULATION

3

3.1 INTRODUCTION

Mathematical models for non-homogeneous populations often become very complex. Therefore a homogeneous population is usually assumed in models for infectious diseases. While this may be adequate for small communities, there are doubts as to its applicability in large populations scattered over wide areas. For instance the transmission of malaria often depends on local ecological factors i.e. it shows spatial dependency. For that reason the assumption of a homogeneous population may be unrealistic for the assessment of the epidemiological course of malaria in a large area. One way out of this problem is to divide the area into two or more sub-areas (or strata), each sub-area being homogeneous and to pool the data collected in these sub-areas.

First a method is given for the comparison of the trend in transmission intensity of two homogeneous populations. It is followed by the main subject:

the assessment of the trend in transmission intensity in a non-homogeneous population. The spatial-dependent and time-dependent transmission model in this chapter can be considered to be an extension of the time-dependent transmission model introduced in the previous chapter. As in chapter 2 it has to be assumed that evidence of exposure to infection is definite for the age band covered by the data.

As an illustration for the application of the model serological data collected in the north of Sri Lanka are used to assess the trend in transmission intensity in the years 1974 - 1978. This trend derived from serological data of one cross-sectional survey carried out in September - October 1978, is compared with the information obtained from malaria surveillance based on the parasitological examination of blood smears during the last five years before survey data in the same area.

3.2 COMPARISON OF THE TREND IN TRANSMISSION INTENSITY OF TWO HOMOGENEOUS POPULATIONS

Two related questions can be asked with regard to the (spline) trend of the transmission intensity in two populations:

- Is the trend in transmission intensity of the two populations equal for all time points t before the survey date
- If the trend of the transmission intensity is not equal in the two populations, which time points t before survey date have different trend values and have different rates of change in the trend of transmission.

In chapter 2 the spline approximations $\lambda_s(t)$ and $\gamma_s(t)$ have been used to describe the trend of the transmission intensity and its rate of change. In this section two populations are considered with $\lambda_{1,s}(t)$, $\gamma_{1,s}(t)$, $\lambda_{2,s}(t)$,

$\gamma_{2,s}(t)$ respectively as parameters for the two populations. The cumulative inoculation rate in age group A_j is indicated by $p_1(A_j)$ and $p_2(A_j)$ respectively ($j=1, \dots, k$).

Overall test for comparing two homogeneous populations

The first question asks whether the null hypothesis $H_0: \lambda_{1,s}(t) = \lambda_{2,s}(t)$ is true for all time points t before survey date. The null hypothesis H_0 implies that the cumulative inoculation rates $p_1(A_j), p_2(A_j)$ are the same for each age group A_j ,

$$p_1(A_j) = p_2(A_j) \quad (j=1, \dots, k),$$

where k is the number of age-groups.

Hence the null hypothesis H_0 can be tested by comparing the cumulative inoculation rates $\hat{p}_1(A_j)$ and $\hat{p}_2(A_j)$ for each age group with the Chi-square test for two proportions (see for instance Everitt (1977)) and by adding the corresponding Chi-square variables.

Hence the test-statistic of the overall test is

$$\chi^2 = \chi_1^2 + \chi_2^2 + \dots + \chi_k^2,$$

where χ_j^2 = Chi-square statistic for the comparison of the two cumulative inoculation rates of age group A_j .

Under the hypothesis H_0 the statistic χ^2 approximately has a Chi-square distribution with k degrees of freedom. If the null hypothesis H_0 is rejected then reference is made to the second question. The proposed tests can be used for a further investigation of the kind of difference between the two populations.

Trend-value test for two homogenous populations

In order to detect time points t before survey date for which the value of the trend in transmission intensity is different in the two populations, for each time point t the hypothesis

$$H_0: \lambda_{1,s}(t) = \lambda_{2,s}(t)$$

is tested against the two-sided alternative

$$H_1: \lambda_{1,s}(t) \neq \lambda_{2,s}(t).$$

Using the large sample normal approximation of the distribution of the test-statistic¹⁾

$$\frac{\hat{\lambda}_{1,s}(t) - \hat{\lambda}_{2,s}(t)}{\sqrt{SD^2 \hat{\lambda}_{1,s}(t) + SD^2 \hat{\lambda}_{2,s}(t)}} \quad (3.1)$$

a two-sided p-value function can be calculated to detect periods before survey date for which the trend-value of the transmission intensity is different in the two populations. The same procedure is followed as in chapter 1: those periods before survey date for which the p-value function takes on a value below the critical value $\alpha = 0.05$ are considered to reflect a difference in transmission level between the two populations.

Trend-change test for two homogeneous populations

An analogous way as described in the trend-value test for two homogeneous populations is followed. For each time point t the null hypothesis

$$H_0: \gamma_{1,s}(t) = \gamma_{2,s}(t) \text{ is tested against the two-sided alternative}$$

$$H_1: \gamma_{1,s}(t) \neq \gamma_{2,s}(t). \text{ The test-statistic is analogous to the test-statistic}$$

1)

$\hat{\lambda}_{i,s}(t)$ = estimate of $\lambda_{i,s}(t)$ in population i ($i=1,2$)

$SD\hat{\lambda}_{i,s}(t)$ = standard deviation of the estimate $\hat{\lambda}_{i,s}(t)$ ($i=1,2$)

used in the trend-value test i.e. $\hat{\lambda}_{i,s}(t)$ is substituted by $\hat{\gamma}_{i,s}(t)$ ($i=1,2$). Under the hypothesis H_0 the distribution of this statistic is asymptotically normal. A difference in the rate of change in the trend of transmission is indicated in those periods for which the p-value function takes on a value below 0.05.

The trend-value test and the trend-change test are introduced here as means to explore the difference between two homogeneous populations indicated by the overall test. However, the trend-value test and the trend-change test can also be applied without performing the overall Chi-square test first. This is particular so if we are interested in the difference between two homogeneous populations with respect to an arbitrary but fixed time point t before survey date.

3.3 SPATIAL-DEPENDENT AND TIME-DEPENDENT TRANSMISSION MODEL

3.3.1 Introduction

In this section measures (parameters) are proposed for the quantitative assessment of the trend of transmission in a non-homogeneous population. Furthermore estimation and testing procedures are presented for the assessment of these parameters. A "*trend-change test*" is introduced to detect a change in the trend of transmission at a time point t years before survey date.

Heterogeneity in a population with respect to the risk of receiving an inoculation may occur within age-classes as well as between them and between subpopulations living in different areas. Here is referred to the spatial variation in the trend of transmission, such as differences between small settlements with regard to the risk of receiving an inoculation. The area

under study is divided into sub-areas and the non-homogeneous population is assumed to consist of homogeneous subpopulations living in these sub-areas. The statistical inference concerns the analysis of cross-sectional data which come from surveys carried out in all subpopulations (fixed design), or from surveys performed only in randomly selected subpopulations (random design).

3.3.2 Parameters for a non-homogeneous population ¹⁾

The average transmission function

To describe the average trend of the transmission intensity in a non-homogeneous population Π , the population is divided into a number of subpopulations Π_i ($i=1, \dots, m$) living in different sub-areas. It is assumed that each subpopulation Π_i ($i=1, \dots, m$) is homogeneous. For a non-homogeneous population the *average transmission function* $\Lambda(t)$ is defined as a weighted linear combination of the transmission functions $\lambda_i(t)$ of the m subpopulations:

$$\Lambda(t) = \sum_{i=1}^m w_i \lambda_i(t) = \text{average transmission function} \quad (3.2)$$

where $\lambda_i(t)$ = transmission function in subpopulation Π_i

w_i = weighting factor for subpopulation Π_i ($\sum w_i = 1$)

In general it is true to say that the value of the average transmission function $\Lambda(t)$ in a time point t represents the mean number of inoculations per annum t years before survey date for an individual randomly chosen from a subpopulation which in turn is randomly selected from the subpopulations with probability w_i for subpopulation Π_i ($i=1, \dots, m$).

There are various possibilities for the selection of a weighting factor

¹⁾ These parameters are denoted by capital letters to distinguish them from the parameters in a homogeneous population.

w_i for subpopulation Π_i ($i=1, \dots, m$) such as:

- a) $w_i = \frac{1}{m}$ (equal weights)
- b) = proportionate to the size of the subpopulation Π_i
- c) = proportionate to the size of sub-area i .

The choice of the weighting factors w_i ($\sum w_i = 1$) determines the value of the average transmission function $\Lambda(t)$. The question which choice has to be preferred depends on the particular objectives of the survey. When it is the objective to study the mean rate of change in transmission intensity of the m subpopulations then alternative a) is chosen ($w_i = \frac{1}{m}$ ($i=1, \dots, m$)). There are situations, however, in which it is more appropriate to take into account the sizes N_i of the subpopulations or the sizes of the areas S_i in which these subpopulations are living ($i=1, \dots, m$).

When it is decided to select alternative b) i.e. w_i is proportionate to the size of subpopulation Π_i

$$w_i = \frac{N_i}{\sum_{i=1}^m N_i} \quad (i=1, \dots, m), \quad (3.3)$$

then $\Lambda(t)$ can be considered an estimate of the mean number of inoculations per annum inflicted on an individual, t years before survey date, who is randomly chosen from the total population.

Generally the sizes N_i ($i=1, \dots, m$) of the m subpopulations change with time and therefore the weighting factors w_i will depend on time t . In this study factors w_i are used which are independent of time.

With regard to alternative c) the weighting factors are

$$w_i = \frac{S_i}{\sum_{i=1}^m S_i} \quad (i=1, \dots, m), \quad (3.4)$$

where S_i is the size of sub-area i .

This means that $\Lambda(t)$ can be interpreted as an estimate of the mean number of inoculations per annum t years before survey date for an individual randomly chosen from those living in a location, which in turn is randomly chosen from all locations of the same size in which the total area can be thought to be divided equally. If the individuals of the total population are uniformly distributed over the total area, i.e. the population density N_i/S_i is constant (independent of i), then weighting proportionate to the sizes of the subpopulations is equivalent with weighting proportionate to the sizes of the sub-areas.

The trend of the average transmission function

As a measure of the trend of the average transmission function $\Lambda(t)$ a weighted linear combination of spline approximations $\lambda_s(t)$ of the sub-populations is used. This *spline approximation of the average transmission function* is denoted by $\Lambda_s(t)$,

$$\Lambda_s(t) = \sum_{i=1}^m w_i \lambda_{i,s}(t). \quad (3.5)$$

The rate of change in the average transmission function $\Lambda(t)$ and its spline approximation $\Lambda_s(t)$ are denoted by $\Gamma(t)$ and $\Gamma_s(t)$ respectively,

$$\Gamma(t) = \frac{d\Lambda(t)}{dt} \quad \text{and} \quad \Gamma_s(t) = \frac{d\Lambda_s(t)}{dt}. \quad (3.6)$$

The spline approximation $\Lambda_s(t)$ is related to the true but unknown average transmission function $\Lambda(t)$ in an analogous way as $\lambda_s(t)$ is related to $\lambda(t)$: the property of *preservation of surface* and the *minimum change* property also hold for a non-homogeneous population. For each interval between two knots there is

$$\int_{T_{j-1}}^{T_j} \Lambda_s(t) dt = \int_{T_{j-1}}^{T_j} \Lambda(t) dt \quad (j=1, \dots, k). \quad (3.7)$$

And $\lambda_s(t)$ is the smoothest function of all those functions which possess the property of preservation of surface and have a continuous first derivative, in formula

$$\int_0^{T_k} \Gamma_s(t)^2 dt \text{ is minimal.} \quad (3.8)$$

For further details see Appendix 3, theorem 3.1.

The average cumulative inoculation rate

Actually it is incorrect to use the spline approximation $\lambda_s(t)$ based on the cumulative inoculation rates $p(A_j)$ of the total population as a measure of the average trend of transmission in a non-homogeneous population. Such an approach might lead to complete misleading results as the following two examples (using hypothetical data) will show.

In the first example (a) we consider a non-homogeneous population of 1120 children (1 to 5 years old). These children are living in two sub-areas; 550 in sub-area 1 (subpopulation 1) and 570 in sub-area 2 (subpopulation 2). It is assumed separately for sub-area 1 and 2 that at any time t the children are exposed to the same risk of receiving an inoculation. The cumulative inoculation rates $p_1(A)$ and $p_2(A)$ in the two respective homogeneous subpopulations and the crude cumulative inoculation rate $p(A)$ in the total population are presented in table 3.1^a (see page 54). The annual trend of the transmission intensity - knots of the spline approximation at time points 0,1,2,3,4,5 years before survey date - in the two subpopulations are presented in fig. 3.1^a (see page 55, solid curves). It is clear that in both subpopulations there has been a downward trend of transmission intensity in a period about 2-5 years before survey date. Hence any quantitative assessment of the average trend of the transmission intensity in the total population (1120 children) should indicate

a decreasing trend in this period before survey date. However, the trend $\lambda_s(t)$ based on the cumulative inoculation rate $p(A)$ of the total population shows just the opposite trend; it is increasing instead of decreasing in the period 2-5 year before survey date.

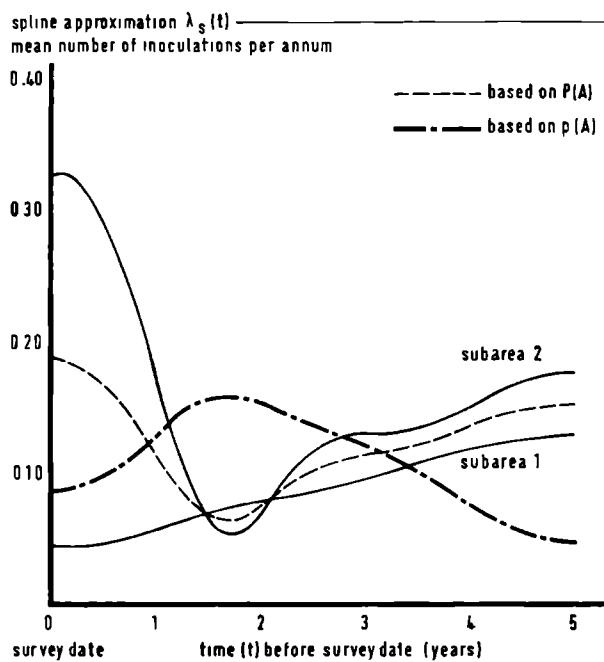
Table 3.1^a Example a; cumulative inoculation rate $p(A)$ versus average¹⁾ cumulative inoculation rate $P(A)$ in a non-homogeneous population (hypothetical data).

age A (years)	Subpopulation 1			Subpopulation 2		
	cumulative inoc. rate (%)			cumulative inoc. rate (%)		
	N_1	N_1^+	$p_1(A)$	N_2	N_2^+	$p_2(A)$
1	150	7	5	50	12	24
2	100	11	11	140	42	30
3	60	11	18	150	56	37
4	90	24	27	150	68	45
5	150	53	35	80	43	54
total	550	106	19	570	221	39

age A (years)	Total population			
	cumulative inoc. rate (%)			average ¹⁾ cumulative inoc. rate $P(A)$ in %
	N	N^+	$p(A)$	
1	200	19	10	15
2	240	53	22	21
3	210	67	32	28
4	240	92	38	37
5	230	96	42	45
total	1120	327	29	30

¹⁾ $1-P(A)$ is the geometric mean of $1-p_1(A)$ and $1-p_2(A)$

Figure 3.1^a Example a ; an artefact in the assessment of the annual trend of transmission in a non-homogeneous population using the time-dependent transmission model: $\lambda_s(t)$ based on the crude cumulative inoculation rate $p(A)$ is misleading.



The question arises how to define the "average" cumulative inoculation rate for a non-homogeneous population in order to apply in a reliable way the time-dependent transmission model¹⁾. The spline function $\lambda_s(t)$ based on this average rate should be a measure of the average trend of transmission. This condition and the definition of the average transmission function $\Lambda(t)$, see formula (3.2), supply the motive to define the average cumulative inoculation rate as follows. *The average cumulative inoculation rate ($P(A)$) is the cumu-*

¹⁾ Of course with appropriate adaptation of sampling errors

lative inoculation rate of a hypothetical homogeneous population exposed to the average transmission intensity ($\Lambda(t)$). Thus by definition

$$P(A) = 1 - e^{- \int_0^A \Lambda(t) dt} \quad (3.9)$$

In appendix 3 it is shown that this definition implies that $P(A)$ is equal to

$$P(A) = 1 - \prod_{i=1}^m (1 - p_i(A))^{w_i}, \quad (3.10)$$

where $p_i(A)$ = cumulative inoculation rate in subpopulation Π_i

w_i = weighting factor for subpopulation Π_i ($\sum w_i = 1$).

The complement of the average inoculation rate ($1-P(A)$) is simply the weighted geometric mean of $1-p_i(A)$ ($i=1, \dots, m$). Using this definition of average cumulative inoculation rate, the non-homogeneous population is formally replaced by hypothetical homogeneous population with cumulative inoculation rate $P(A)$.

The transmission function $\lambda(t)$ and its spline approximation $\lambda_s(t)$ based on $P(\Lambda_j)$ ($j=1, \dots, k$) then represent the average transmission function $\Lambda(t)$ and its spline approximation $\Lambda_s(t)$ respectively. The data of the first example (table 3.1^a) are used to illustrate this. $P(A)$ is calculated with equal weights for the two subpopulations ($w_1 = w_2 = \frac{1}{2}$). The spline trend $\lambda_s(t)$ based on $P(\Lambda_j)$ ($j=1, 2, 3, 4, 5$) is shown in fig. 3.1^a. This figure clearly shows that $\lambda_s(t)$ based on the average cumulative inoculation rate measures the (unweighted) mean trend of the transmission intensity in the two populations.

Eventual artefacts in the assessment of the average trend of the transmission intensity in a non-homogeneous population, using the cumulative inoculation rates of the total population, does not only concern a possible false upward trend in transmission intensity. A false downward trend may occur in the entire period as the second example (b) will show. In table

3.1^b a non-homogeneous population of 895 children is considered; 491 are living in sub-area 1 and 404 are living in sub-area 2. If it is assumed that homogeneous subpopulations are under study then the cumulative inoculation rates $p_1(A)$ and $p_2(A)$ in table 3.1^b are a result of transmission intensities $\lambda_1(t)$ and $\lambda_2(t)$ respectively which are both increasing with time.

Table 3.1^b Example b; cumulative inoculation rate $p(A)$ versus average¹⁾ cumulative inoculation rate $P(A)$ in a non-homogeneous population (hypothetical data).

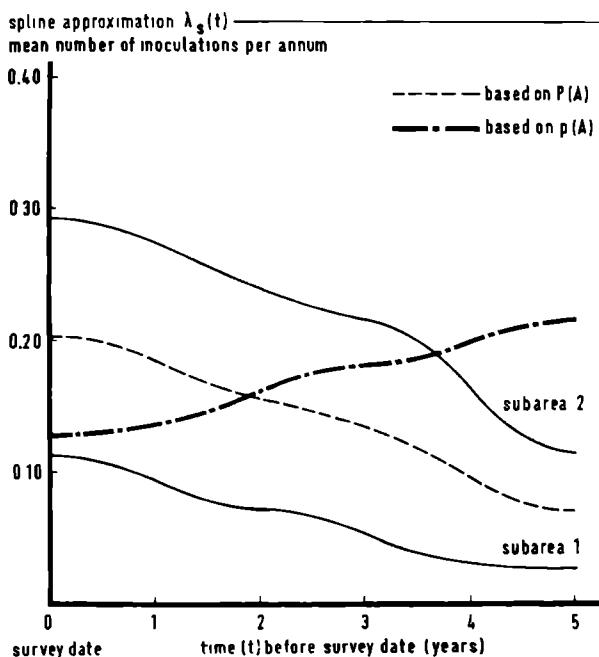
age A (years)	Subpopulation 1			Subpopulation 2		
	cumulative inoc. rate (%)			cumulative inoc. rate (%)		
	N_1	N_1^+	$p_1(A)$	N_2	N_2^+	$p_2(A)$
1	150	15	10	24	6	25
2	125	21	17	50	21	42
3	100	22	22	80	43	54
4	68	17	25	100	62	62
5	48	13	27	150	100	67
total	491	88	18	404	232	57

age A (years)	Total population			
	cumulative inoc. rate (%)			average ¹⁾ cumulative
	N	N^+	$p(A)$	inoc. rate $P(A)$ in %
1	174	21	12	18
2	175	42	24	31
3	180	65	36	40
4	168	79	47	47
5	198	113	57	51
total	895	320	36	41

¹⁾ $1-P(A)$ is the geometric mean of $1-p_1(A)$ and $1-p_2(A)$

The annual trends $\lambda_{1,s}(t)$ and $\lambda_{2,s}(t)$ are presented in fig. 3.1^b, they both show an upward trend in transmission for the entire period under study. However the spline function $\lambda_s(t)$ based on the cumulative inoculation rate $p(A)$ in the total population shows a downward trend. This artefact is the result of ignoring the fact that a non-homogeneous population was under consideration. Differences in sizes of age-groups might then contribute to a completely misleading picture. However, when the average cumulative inoculation rates $P(A_j)$ ($j=1,\dots,k$) are used (see table 3.1^b), such an artefact cannot occur.

Figure 3.1^b Example b; an artefact in the assessment of the annual trend of transmission in a non-homogeneous population using the time-dependent transmission model: $\lambda_s(t)$ based on the crude cumulative inoculation rate $p(A)$ is misleading.



The spline function $\lambda_s(t)$ based on the average cumulative inoculation rate $P(A)$ with equal weightage for the two subpopulations presents the (unweighted) mean trend of transmission in the two populations, see fig. 3.1.^b.

In the two examples considered here the spline trend $\lambda_s(t)$ based on the cumulative inoculation rate $p(A)$ in the total population is completely contradictory to the spline trend $\lambda_s(t)$ based on the average cumulative inoculation rate $P(A)$ which is the true annual trend. These examples are of course far-fetched and influenced by the sizes of the age-groups, however they clearly show which the pitfalls are if a non-homogeneous population is considered as homogeneous.

Spatial variation

As a measure of the spatial variation in the trend of the transmission intensity and the spatial variation in its rate of change, the weighted variance of $\lambda_{i,s}(t)$ and $\gamma_{i,s}(t)$ are used respectively. These variances are denoted by $\sigma^2(\lambda_s(t))$ and $\sigma^2(\gamma_s(t))$. Thus by definition

$$\begin{aligned}\sigma^2(\lambda_s(t)) &= \sum_{i=1}^m w_i [\lambda_{i,s}(t) - \lambda_s(t)]^2 \\ &= \text{spatial variance in } \lambda_s(t).\end{aligned}\quad (3.11)$$

The formula of $\sigma^2(\gamma_s(t))$ is completely analogous to the formula of $\sigma^2(\lambda_s(t))$.

The emphasis so far in this section has been on the definition of parameters for a non-homogeneous population. The statistical inference concerning these parameters is presented in the next section. This inference mainly concerns the spline functions $\lambda_{i,s}(t)$, $\gamma_{i,s}(t)$ (homogeneous subpopulations), $\lambda_s(t)$, $\gamma_s(t)$ (non-homogeneous population) and the parameters $\sigma^2(\lambda_s(t))$, $\sigma^2(\gamma_s(t))$ respectively. Therefore the clarity of the notation in the next

section will clearly improve if the parameter t (time before survey date) and the subscript s (spline approximation) can be suppressed in the formulae of the parameters. Accordingly the notation λ_i , γ_i , Λ , Γ and σ_λ^2 , σ_γ^2 will often be used rather than the more exact but complicated notation.

3.3.3 Statistical inference, trend-change test

3.3.3.1 Introduction

In this section estimators of the spline-trend $\Lambda_s(t)$ and its rate of change $\Gamma_s(t)$ are presented. Formulae are given for the assessment of the spatial variation in the trend of the transmission intensity using analysis of variance concepts. Furthermore a statistical test is proposed for the detection of a change in the average trend of transmission.

Two sample designs are considered namely, the fixed design (F-design) and the random design (R-design). In an F-design, cross-sectional surveys are carried out in all subpopulations Π_i ($i=1, \dots, m$) of the non-homogeneous population under study. In an R-design, cross-sectional surveys are only performed in a (weighted) *random sample* of subpopulations (n) chosen from all subpopulations (m) into which the non-homogeneous population has been divided. The subpopulations in which the surveys are actually carried out are denoted by $P_{i_1}, P_{i_2}, \dots, P_{i_n}$. In the applications an analysis can be based on an F-design as well as on an R-design. A comparison is made between these two approaches of data analysis.

Before applying an analysis based on an F-design and an R-design respectively it is recommended to perform an overall test for the detection of spatial variation in the trend of transmission. If cross-sectional serological surveys are carried out in n homogeneous subpopulations (n sub-areas) then the

following test can be applied:

Overall test for the detection of spatial variation

To test the hypothesis "no spatial variation" a simple generalization of the overall test for comparing two homogeneous populations is used, see section 3.2. No spatial variation implies that the following null hypothesis is true:

$$H_0: \lambda_{1,s}(t) = \lambda_{2,s}(t) = \dots = \lambda_{n,s}(t) \text{ for all time points } t \quad (0 \leq t \leq T_k).$$

To test this hypothesis the statistic $\chi^2 = \sum_{j=1}^k \chi_j^2$ is used; with χ_j^2 is the Chi-square statistic for the comparison of the n cumulative inoculation rates of age group A_j (null hypothesis: $p_1(A_j) = p_2(A_j) = \dots = p_n(A_j)$). Under H_0 the distribution of the statistic χ_j^2 has a Chi-square distribution with $(n-1)$ degrees of freedom¹⁾ and thus (under H_0) χ^2 is a Chi-square variable with $k(n-1)$ degrees of freedom.

If the null hypothesis is not rejected in sufficiently large investigations i.e. spatial variation is not detectable with this test, then the data of the n subpopulations can be put together (by age). The combination of the n subpopulations is then considered as homogeneous. This would enable the time-dependent transmission model and the related statistical inference as described in chapter 2 to be applied. On the other hand if there is an indication of spatial variation then one should perform an analysis based on the statistical inference presented in this section (F-design and/or R-design).

Conventions with regard to the notation of estimators (estimates)

The simplified notation λ_i , γ_i , Λ , Γ and σ_λ^2 , σ_γ^2 will often be used to

¹⁾ see for instance Everitt (1977)

indicate parameters (see section 3.3.2). The estimators (estimates) of parameters are indicated generally by a " $\hat{\cdot}$ " above the parameters. Accordingly they are denoted by $\hat{\lambda}_i$, $\hat{\gamma}_i$, $\hat{\Lambda}$, $\hat{\Gamma}$ and $\hat{\sigma}_{\lambda}^2$, $\hat{\sigma}_{\gamma}^2$ respectively. Estimators are derived both in a fixed design and a random design. To indicate the design the underscripts F and R are used; e.g. $\hat{\Lambda}_F$ is the estimator (estimate) of Λ in a fixed design.

The standard deviations¹⁾ of estimators (estimates) are denoted by SD(); for example $SD(\hat{\Lambda}_F)$ is the standard deviation of $\hat{\Lambda}_F$ in an F-design.

As the estimators of Γ and σ_{γ}^2 can be defined and derived in an analogous way as the estimators of Λ and σ_{λ}^2 , formulae for $\hat{\Lambda}$, its standard deviation $SD\hat{\Lambda}$ and $\hat{\sigma}_{\lambda}^2$ are only presented; the expressions for $\hat{\Gamma}$, $SD\hat{\Gamma}$ and $\hat{\sigma}_{\gamma}^2$ are completely similar to these.

3.3.3.2 Fixed design

A fixed design is based on the stratification of the population in m homogeneous subpopulations living in m sub-areas. Cross-sectional serological surveys are carried out in all subpopulations. Hence for each subpopulation Π_i ($i=1, \dots, m$) an estimator of the spline trend of the transmission intensity $\lambda_s(t)$ is available. As explained in the introduction this estimator is now abbreviated as $\hat{\lambda}_i$. In an F-design the estimator of the spline approximation of the average transmission function is denoted by $\hat{\Lambda}_F$, its standard deviation¹⁾ is denoted as $SD\hat{\Lambda}_F$,

$$\hat{\Lambda}_F = \sum_{i=1}^m w_i \hat{\lambda}_i \quad SD^2 \hat{\Lambda}_F = \sum_{i=1}^m w_i^2 SD^2 \hat{\lambda}_i. \quad (3.12)$$

For the estimation of the spatial variation in the trend values λ_i ($i=1, \dots, m$) (the parameter σ_{λ}^2) the following estimator denoted by $\hat{\sigma}_{F,\lambda}^2$ is used

¹⁾ Note, here is referred to estimates of standard deviations of estimators

$$\hat{\sigma}_{F,\lambda}^2 = \sum_{i=1}^m w_i (\hat{\lambda}_i - \hat{\lambda}_F)^2 - \frac{1}{\sum_i w_i} \sum_{i=1}^m w_i (1-w_i) S D^2 \hat{\lambda}_i. \quad (3.13)$$

The expression on the right side of (3.13) may take on negative values. This would imply that the variation in the spline approximations $\hat{\lambda}_i$ ($i=1, \dots, m$) is completely explained by the statistical errors in these spline approximations $S D \hat{\lambda}_i$ ($i=1, \dots, m$), which are due to the sampling of individuals in the subpopulations. The question arises as to which part of the variation in $\hat{\lambda}_i$ can be ascribed to sampling errors and which part of the variation in $\hat{\lambda}_i$ is a result of the spatial variation in the trend of the transmission intensity. To determine the relative influence of these two sources of variation on the total variation in $\hat{\lambda}_i$ ($i=1, \dots, m$), the following descriptive definitions of "total variance", "spatial variance" and "error variance" are used (see (3.13)):

$$V_{F,tot} = \text{total variance} = \sum_{i=1}^m w_i (\hat{\lambda}_i - \hat{\lambda}_F)^2 \quad (3.14)$$

$$V_{F,spat} = \text{spatial variance} = \hat{\sigma}_{F,\lambda}^2 \quad (3.15)$$

$$V_{F,err} = \text{error variance} = \sum_{i=1}^m w_i (1-w_i) S D^2 \hat{\lambda}_i. \quad (3.16)$$

Using this convention the total variance $V_{F,tot}$ can be written as the sum of the spatial variance $V_{F,spat}$ and the error variance $V_{F,err}$,

$$V_{F,tot} = V_{F,spat} + V_{F,err}. \quad (3.17)$$

That part of the total variance that can be explained by statistical fluctuations due to the sampling of individuals is called proportion explained variance, notation E_F ,

$$E_F = \frac{V_{F,err}}{V_{F,tot}}. \quad (3.18)$$

As the error variance may be larger than the total variance the proportion explained variance E_F may assume a value > 1 .

For the estimation of Γ and σ^2_Y estimators $\hat{\Gamma}_F$ and $\hat{\sigma}^2_{F,Y}$ are used and these are completely similar to $\hat{\Lambda}_F$ and $\hat{\sigma}^2_{F,\lambda}$ respectively. (In the formulae of $\hat{\Lambda}_F$, $SD^2\hat{\Lambda}_F$ and $\hat{\sigma}^2_{F,\lambda}$ the $\hat{\lambda}_i$ has to be replaced by $\hat{\gamma}_i$ ($i=1, \dots, m$)). Furthermore the total variance, the spatial variance the error variance and the proportion explained variance of $\hat{\gamma}_i$ ($i=1, \dots, m$) are defined by formulae completely analogous to the formulae presented above.

Trend-change test for a non-homogeneous population (F-design)

For the detection of changes in the spline trend $\Lambda_s(t)$ a method similar to the method described for a homogeneous population is proposed (see chapter 2). For each time point t before survey date the following hypothesis H_0 is tested against the two-sided alternative H_1 :

H_0 : the rate of change in $\Lambda_s(t)$ is zero

H_1 : the trend function $\Lambda_s(t)$ is increasing or decreasing.

To test the hypothesis H_0 the statistic $T_F(t)$ is used (*trend-change test*),

$$T_F(t) = \frac{\hat{\Gamma}_F}{SD\hat{\Gamma}_F} . \quad (3.19)$$

with $\hat{\Gamma}_F$ and $SD\hat{\Gamma}_F$ defined by (see (3.12))

$$\hat{\Gamma}_F = \sum_{i=1}^m w_i \hat{\gamma}_i \text{ and } SD\hat{\Gamma}_F = \sqrt{\sum_{i=1}^m w_i^2 SD^2\hat{\gamma}_i} . \quad (3.20)$$

Using the large sample normal approximation of the distribution of $\hat{\Gamma}_F(t)$ a two-sided p-value is calculated for each time point t before survey date. Those time points in which the p-value function, notated as $P_F(t)$, takes on values less than the critical value $\alpha=0.05$, are considered to reflect a change

in the average trend function $\Lambda_s(t)$.

It must be emphasized that the meaning of the trend function $\Lambda_s(t)$ and therefore the meaning of the null hypothesis H_0 depends on the weighting factors w_i ($i=1, \dots, m$). Hence a particular choice of these factors implies a particular null hypothesis H_0 and each choice of w_i ($i=1, \dots, m$) corresponds with a different test-statistic $T_F(t)$ and different p-value function $P_F(t)$.

If $\gamma_1 = \gamma_2 = \dots = \gamma_m = 0$ then $\Gamma = \sum_{i=1}^m w_i \gamma_i = 0$ for any choice of the weighting factors w_i ($i=1, \dots, m$). Therefore each test-statistic $T_F(t)$ - each choice of w_i - may in principle be used for testing the hypothesis: "The trend in transmission intensity did not change t years before survey date in all subpopulations". Under this null hypothesis ($\gamma_1 = \gamma_2 = \dots = \gamma_m = 0$) the statistic $T_F(t)$ will be distributed approximately as a normal variable for any choice of the weighting factors w_i ($i=1, \dots, m$). The power of the test $T_F(t)$, however, depends on these weighting factors and also on the unknown γ_i ($i=1, \dots, m$). As a simple measure of this power the following expression can be used¹⁾

$$q_1 = \frac{\sum_{i=1}^m w_i \gamma_i}{\sqrt{\sum_{i=1}^m w_i^2 \text{Var } \hat{\gamma}_i}} . \quad (3.21)$$

Two alternatives for the trend-change test $T_p(t)$

A priori the γ_i are of course unknown. However, it is possible to have some knowledge about the values $\text{Var } \hat{\gamma}_i$ ($i=1, \dots, m$). For instance, if the sample sizes of the surveys in the m subpopulations are widely different then it can be expected that sampling in certain subpopulations may have a relatively large influence on the power of the trend-change test (see 3.21). In

¹⁾ γ_i = the rate of change of $\lambda_s(t)$ in subpopulation Π_i ; $\text{Var } \hat{\gamma}_i$ is the variance of the estimator $\hat{\gamma}_i$

such a situation it might be preferable to use a test based on the sum of the standardized rates of change of $\hat{\lambda}_i$ ($i=1, \dots, m$), i.e.

$$\frac{1}{\sqrt{m}} \sum_{i=1}^m \frac{\hat{\gamma}_i}{\text{SD}(\hat{\gamma}_i)} . \quad (3.22)$$

Under the null hypothesis "all $\gamma_i = 0$ " this statistic is asymptotically distributed as a standardized normal variable. The power of such a test can be measured by

$$q_2 = \frac{1}{\sqrt{m}} \sum_{i=1}^m \frac{\gamma_i}{\sqrt{\text{Var} \hat{\gamma}_i}} . \quad (3.23)$$

If the sample size in one subpopulation n_i is extremely small and therefore $\text{Var} \hat{\gamma}_i$ relatively very high, then this may have a relatively large effect on the power q_1 (3.21) as compared with its effect on q_2 (3.23). The test based on the sum of the standardized rates of change may then be more suitable to detect changes in the trend of the transmission intensity than the trend-change test $T_p(t)$ with equal weightage for the subpopulations. A more sophisticated approach for determining weighting factors would take into account the views presented in section 3.3.2 as well as the statistical considerations briefly mentioned here.

Another method occasionally used to compound all the evidence obtained from statistical data, is the method of combination of p-values of several mutually independent tests (introduced by *Fisher* (1944)). This method may be applied here to pool the one-sided p-value functions $p_i(t)$ obtained from the subpopulations (see chapter 2). For each time point t before survey date the statistic

$$- \sum_{i=1}^m 2^e \log p_i(t) \quad (3.24)$$

is used to test the null hypothesis "all $\gamma_i = 0$ ". Under the null hypothesis

this statistic has asymptotically a Chi-square distribution with $2m$ degrees of freedom.

Tests may be combined in several other ways than described here, and each combination will lead to a different result as each is based on a different function of the observations. The choice among the different possible combinations should be made on account of the alternative hypothesis. It may, however, be difficult to solve this problem, and in such cases Fisher's "omnibus" - test may be used, even if it is not the most powerful (Hald, 1957).

In the following mainly the trend-change test $T_F(t)$ will be used for testing the hypothesis "*the rate of change in $\Lambda_s(t)$ is zero*". For purpose of comparison however, the two other tests described above are also applied in this chapter. A significant test result in a time point t years before survey date ($p \leq 0.05$, one-sided) for these tests is interpreted as "*the (spline-) trend of transmission intensity has been increasing (decreasing) in at least one subpopulation*".

3.3.3.3 Random design

In a random design, notation R-design, a certain number (n) of sub-areas are "randomly" selected from the m sub-areas into which the total area has been divided. In order to obtain a weighted random sample of subpopulations, n independent observations are taken of the random variable I defined by

$$P[I=i] = w_i \quad (i=1, \dots, m),$$

where w_i are the weighting factors described in section 3.3.2 ($\sum w_i = 1$).

The observations of the variable I are denoted by i_1, i_2, \dots, i_n .

The cross-sectional surveys are actually carried out in the subpopulations

(sub-areas) $\Pi_{i_1}, \Pi_{i_2}, \dots, \Pi_{i_n}$. It is assumed that either the selection of the

sub-areas has been done with replacement or when the sampling of sub-areas has been done without replacement that n is relatively small as compared with m .

In an R-design the inference is made on trend functions which are stochastic itself. The random spline approximation associated with λ_{ij} and its estimator with standard deviation are denoted as $\hat{\lambda}_{ij}$, $\hat{\lambda}_{ij}$ and $SD^2\hat{\lambda}_{ij}$ ($j=1, \dots, n$) respectively. For the estimation of the average trend function the following estimator denoted by $\hat{\Lambda}_R$ is used (its standard deviation is denoted as $SD\hat{\Lambda}_R$),

$$\hat{\Lambda}_R = \frac{1}{n} \sum_{j=1}^n \hat{\lambda}_{ij} \quad SD^2\hat{\Lambda}_R = \frac{1}{n(n-1)} \sum_{j=1}^n (\hat{\lambda}_{ij} - \hat{\Lambda}_R)^2. \quad (3.25)$$

The spatial variation in the trend values λ_i in the m subpopulations i.e. the parameter σ_λ^2 is estimated by

$$\hat{\sigma}_{R,\lambda}^2 = \frac{1}{n-1} \sum_{j=1}^n (\hat{\lambda}_{ij} - \hat{\Lambda}_R)^2 - \frac{1}{n} \sum_{j=1}^n SD^2\hat{\lambda}_{ij}. \quad (3.26)$$

In an R-design the variation of the spline approximations $\hat{\lambda}_{ij}$ ($j=1, \dots, n$) is the result of the influence of three factors: the spatial variation in the trend of the transmission intensity as described by σ_λ^2 , the statistical fluctuations due to the (weighted) random sampling of n spline approximations λ_{ij} ($j=1, \dots, n$) and the statistical errors in the estimates $\hat{\lambda}_{ij}$ due to the sampling of individuals. To study that part of the variation in $\hat{\lambda}_{ij}$ ($j=1, \dots, n$) that can be explained by the sampling of subpopulations and the sampling of individuals in these subpopulations, concepts similar to the concepts defined in the previous section are used. For an R-design the definitions are:

$$V_{R,tot} = \text{total variance} = \frac{1}{n-1} \sum_{i=1}^n (\hat{\lambda}_{ij} - \hat{\Lambda}_R)^2, \quad (3.27)$$

$$V_{R,spat} = \text{spatial variance} = \hat{\sigma}_{R,\lambda}^2, \quad (3.28)$$

$$V_{R,err} = \text{error variance} = \frac{1}{n} \sum_{i=1}^n SD^2 \hat{\lambda}_{ij}. \quad (3.29)$$

Using this convention the total variance is split up into two components,

$$V_{R,tot} = V_{R,spat} + V_{R,err}. \quad (3.30)$$

The proportion explained variance due to sampling is similarly defined as in the F-design, that is

$$E_R = \frac{V_{R,err}}{V_{R,tot}}. \quad (3.31)$$

As the error variance $V_{R,err}$ might be larger than the total variance $V_{R,tot}$, the estimate of the spatial variation in the trend values $V_{R,spat}$ may take on negative values and hence the proportion explained variance E_R may assume a value > 1 .

For the estimation of γ and σ^2_γ and the description of the sources of variation in $\hat{\gamma}_{ij}$ ($j=1, \dots, n$) exactly the same procedure is followed as described above.

Trend-change test for a non-homogeneous population (R-design)

In order to detect a change in $\Lambda_s(t)$ an analogous way is followed as described for a fixed design (see previous section). For each time point t before survey date the hypothesis H_0 "the rate of change in $\Lambda_s(t)$ is zero" is tested against the two-sided alternative H_1 "the trend function $\Lambda_s(t)$ is increasing or decreasing" (the trend-change test). The test-statistic used is

$$T_R(t) = \frac{\hat{\Gamma}_R}{SD\hat{\Gamma}_R}, \quad (3.32)$$

with \hat{r}_R and SD \hat{r}_R defined by (see (3.25))

$$\hat{r}_R = \frac{1}{n} \sum_{j=1}^n \hat{\gamma}_{ij} \text{ and } \text{SD} \hat{r}_R = \sqrt{\frac{1}{n(n-1)} \sum_{j=1}^n (\hat{\gamma}_{ij} - \bar{\hat{r}}_R)^2}. \quad (3.33)$$

Using the large sample normal approximation of the distribution of $T_R(t)$ a two-sided p-value function, notated as $P_R(t)$, is calculated. Indicative for a change in the spline approximation $\Lambda_s(t)$ of the average transmission function are those periods for which $P_R(t) \leq 0.05$.

The emphasis placed on the meaning of $\Lambda_s(t)$, the weighting factors w_i ($i=1, \dots, m$) and the null hypothesis H_0 in an F-design, equally holds for the R-design. The test based on the sum of the standardized rates of change $\hat{\gamma}_{ij}$ ($j=1, \dots, n$) and the test based on the pooling of p-value functions described in the previous section may also be used for testing the null hypothesis " $\gamma_i = 0$ ($i=1, \dots, m$)" in an R-design. Under this null hypothesis the distributions of the test-statistics of these two tests are independent of the method used to select sub-areas. For the application of the two tests it is therefore not required that the n sub-areas are randomly selected from the m sub-areas into which the total area has been divided. However, when there is a large spatial variation in the trend of transmission then the power of these two tests to reject the hypothesis " $\gamma_i = 0$ " might of course be strongly dependent on the particular method to select sub-areas.

3.3.3.4 Relationship between the analyses based on an F-design and on an R-design

When cross-sectional surveys are carried out in n homogeneous subpopulations Π_1, \dots, Π_n , then an analysis based on an F-design as well as one based on an R-design might be performed. When the analysis is

based on the theory of an F-design then the conclusions of the analysis are restricted to the population which consists of the compounding of these n sub-populations (notated as $\bigcup_{i=1}^n \Pi_i$). If it is acceptable to assume that the sub-populations Π_1, \dots, Π_n are a random sample (with replacement) from m homogeneous subpopulations - i.e. the non-homogeneous population Π under study - then an analysis based on an R-design is justified. The conclusions of the analysis in such a design are then valid for a much larger population (i.e. $\bigcup_{i=1}^m \Pi_i$) than the population in which the surveys have been actually carried out ($\bigcup_{i=1}^n \Pi_i$).

In practical applications it will not often occur that the "sampling" of subpopulations Π_1, \dots, Π_n has been performed with appropriate weighting factors w_i ($i=1, \dots, m$). In the planning of a survey the selection of sub-areas among other things is restricted by administrative, technical and logistic problems. In such a situation it might even be difficult to argue that the sample of subpopulations Π_1, \dots, Π_n can be regarded effectively as a random sample with equal weightage. In the following a comparison is made of an analysis based on an F-design with one based on an R-design assuming equal weightage for the sub-populations in each design.

In case of an F-design an assessment is made of the average trend function and the spatial variation in the population $\bigcup_{i=1}^n \Pi_i$. These parameters are denoted by Λ_F and $\sigma_{F,\lambda}^2$, respectively,

$$\Lambda_F = \frac{1}{n} \sum_{i=1}^n \lambda_i \text{ and } \sigma_{F,\lambda}^2 = \frac{1}{n} \sum_{i=1}^n (\lambda_i - \Lambda_F)^2. \quad (3.34)$$

The statistical inference based on an R-design concerns the average trend function and the spatial variation in the population $\bigcup_{i=1}^m \Pi_i$, notated as Λ_R and $\sigma_{R,\lambda}^2$,

$$\Lambda_R = \frac{1}{m} \sum_{i=1}^m \lambda_i \text{ and } \sigma_{R,\lambda}^2 = \frac{1}{m} \sum_{i=1}^m (\lambda_i - \Lambda_R)^2. \quad (3.35)$$

Emphasis is placed on the difference between

$\hat{\Lambda}_F$ and $\hat{\Lambda}_R$, $\sigma_{F,\lambda}^2$ and $\sigma_{R,\lambda}^2$ respectively.

The estimate of $\hat{\Lambda}_F$ with standard deviation and the estimate of $\hat{\Lambda}_R$ with standard deviation are (see formulae (3.12) with $m=n$, $w_i=1/n$ and formula (3.25) with $\hat{\lambda}_{ij}$ substituted by $\hat{\lambda}_i$)

$$\hat{\Lambda}_F = \frac{1}{n} \sum_{i=1}^n \hat{\lambda}_i \quad SD\hat{\Lambda}_F = \frac{1}{n} \sqrt{\sum_{i=1}^n SD^2 \hat{\lambda}_i} \quad (3.36)$$

$$\hat{\Lambda}_R = \frac{1}{n} \sum_{j=1}^n \hat{\lambda}_i \quad SD\hat{\Lambda}_R = \sqrt{\frac{1}{n(n-1)} \sum_{i=1}^n (\hat{\lambda}_i - \hat{\Lambda}_R)^2}. \quad (3.37)$$

Although the estimates $\hat{\Lambda}_F$ and $\hat{\Lambda}_R$ are identical their standard deviations are completely different. Note: the standard deviation in an R-design ($SD\hat{\Lambda}_R$) can be smaller than the standard deviation in an F-design ($SD\hat{\Lambda}_F$). The estimates of $\sigma_{F,\lambda}^2$ and $\sigma_{R,\lambda}^2$ are

$$\hat{\sigma}_{F,\lambda}^2 = \frac{1}{n} \sum_{i=1}^n (\hat{\lambda}_i - \hat{\Lambda}_F)^2 - \frac{n-1}{n^2} \sum_{i=1}^n SD^2 \hat{\lambda}_i \quad (3.38)$$

$$\hat{\sigma}_{R,\lambda}^2 = \frac{1}{n-1} \sum_{i=1}^n (\hat{\lambda}_i - \hat{\Lambda}_R)^2 - \frac{1}{n} \sum_{i=1}^n SD^2 \hat{\lambda}_i. \quad (3.39)$$

It is easily seen from (3.38) and (3.39) that the estimate of the spatial variation in the large population ($\sum_{i=1}^m \Pi_i$) is $n/(n-1)$ times larger than the estimate of the spatial variation in the population actually examined

$$(\sum_{i=1}^n \Pi_i),$$

$$\hat{\sigma}_{R,\lambda}^2 = \frac{n}{n-1} \hat{\sigma}_{F,\lambda}^2. \quad (3.40)$$

As the parameters Γ_F , $\sigma_{F,Y}^2$ and Γ_R , $\sigma_{R,Y}^2$ are defined in an analogous way

as Λ_F , $\sigma_{F,\lambda}^2$ and Λ_R , $\sigma_{R,\lambda}^2$ respectively, it follows immediately that similar estimators and standard deviations can be derived for these parameters.

Of special interest are the total variance and the error variance of $\hat{\gamma}_i$ ($i=1, \dots, n$). In an F-design these concepts are given by (see (3.14) and (3.16) with $m=n$ and $w_i = \frac{1}{n}$),

$$V_{F,tot} = \frac{1}{n} \sum_{i=1}^n (\hat{\gamma}_i - \bar{\gamma}_F)^2 \quad V_{F,err} = \frac{n-1}{n^2} \sum_{i=1}^n SD^2 \hat{\gamma}_i. \quad (3.41)$$

From the definition of the total variance and error variance in an R-design it follows that, see (3.27) and (3.29),

$$V_{R,tot} = \frac{n}{n-1} V_{F,tot} \quad V_{R,err} = \frac{n}{n-1} V_{F,err}. \quad (3.42)$$

The proportion explained variance in an F-design (E_F) as well as in an R-design (E_R) has been defined as the ratio of the error variance and the total variance. It follows immediately that $E_F = E_R$. Hence when equal weightage is used for the subpopulations then the proportion explained variance in the rates of change $\hat{\gamma}_i$ is equal for both designs. This proportion is therefore notated as E. Generally E depends on time t, thus

$$E(t) = E_F(t) = E_R(t). \quad (3.43)$$

When the sample sizes in the subpopulations π_1, \dots, π_n become very large then one expects the error variance to become smaller and in general a smaller fraction of the total variance is explained by sampling errors. On the other hand, when the sample sizes in the subpopulations are relatively small, then the error variance is relatively large, it may even occur that the error variance becomes larger than the total variance, i.e. $E(t) > 1$, indicating that the variation in the change of the trend in transmission intensity is

completely explained by the statistical fluctuations due to the relatively small samples. An adequate assessment of the spatial variation in the rate of change γ_i would then require larger samples for serological examination.

There exists a simple relationship between the proportion explained variance $E(t)$ for the variation in $\hat{\gamma}_i$ and the test-statistics $T_F(t)$ and $T_R(t)$ presented in section 3.3.3.2 and 3.3.3.3. respectively. In general the following relation holds

$$\frac{T_R(t)}{T_F(t)} = \sqrt{E(t)}. \quad (3.44)$$

As a consequence of (3.44) the following straight-forward relationship exists between the proportion explained variance in $\hat{\gamma}_i$ and the p-value functions in the F-design and R-design:

$$\begin{aligned} E(t) < 1 &\Leftrightarrow P_R(t) > P_F(t) \\ E(t) = 1 &\Leftrightarrow P_R(t) = P_F(t) \\ E(t) > 1 &\Leftrightarrow P_R(t) < P_F(t) \end{aligned} \quad (3.45)$$

where: $E(t)$ = proportion explained variance of the total variance in $\hat{\gamma}_i$
 $(i=1,\dots,n)$ due to the sampling of individuals;

$P_F(t)$ = p-value function for the detection of change in the average trend function $\Lambda_F(t)$ in the combined subpopulations Π_1, \dots, Π_n ;
 $P_R(t)$ = p-value function for the detection of change in the average trend function $\Lambda_R(t)$ in the combined subpopulations Π_1, \dots, Π_m .

Hence, when the variation in the rate of change $\hat{\gamma}_i$ is "over explained" by sampling errors, that is $E(t) > 1$, then the p-value function for the detection of a change in the trend function Λ_R takes on a smaller value than the p-value function for the detection of a change in Λ_F . It might even occur that at time t there is a significant change in trend of transmission in the large

population $\sum_{i=1}^m P_i$ ($P_R(t) \leq 0.05$), which is not reflected in the restricted population $\sum_{i=1}^n P_i$ ($P_F(t) > 0.05$).

3.4 ILLUSTRATION, USING DATA COLLECTED IN SRI LANKA

3.4.1 Introduction

Malaria in Sri Lanka is of an unstable nature. Malaria control is given high priority by the government of the island. This implies also that there is a great need for an effective evaluation of the malaria control operations.

The vector of malaria in Sri Lanka, *Anopheles culicifacies*, was found to be increasingly resistant to DDT; therefore this insecticide had to be replaced. In Kilinochchi, an area in the north of Sri Lanka, DDT was replaced by malathion in September–October 1977. A downward trend in transmission intensity following the introduction of this new insecticide would provide an indication for the effectiveness of malathion in this area.

In order to assess the trend in transmission intensity in Kilinochchi a study has been undertaken by the University of Colombo, the Anti Malaria Campaign (AMC) of Sri Lanka¹⁾ and the University of Nijmegen²⁾ (The Netherlands). The specific objectives were: a) the assessment of the trend in transmission intensity in the last five years before Sept/Oct 1978 and b) to test the validity of the mathematical-statistical models developed in this study.

The results of the application of the spatial-time dependent transmission model are presented here (sample size 1678). These results are compared with

¹⁾ University of Colombo, Faculty of Medicine, Dept. of Parasitology (Dr. S. Rajakulendran); Anti Malaria Campaign (Dr. K. Subramaniam).

²⁾ University of Nijmegen, Faculty of Medicine, Dept. of Parasitology (Dr. J.H.E.Th. Meuwissen, Dr. T. Ponnudurai).

the trend in transmission intensity derived from parasitological data collected by the AMC in the period 1973-1978 in the same area (examination of 14,943 blood smears).

3.4.2 The design of the survey

The Kilinochchi area has been selected as a study area for various reasons. It was necessary to choose an area with a sufficiently large population and a fairly high incidence of malaria. Obtaining the necessary sample size was essential for the success of the study. It was also necessary to see that the spraying operations and the collection of serological and parasitological data was adequately supervised. These prerequisites seemed to be assured in Kilinochchi an area in the north of Sri Lanka. The population size of the area is 104,000. It is one of the Health Areas demarcated by the Anti Malaria Campaign (AMC) of Sri Lanka in which an integrated malaria control programme is being carried out. This is implemented by the endoor spraying of malathion at a concentration of 2 gms/square meter in a three monthly cycle. Since 1978 a coverage of 80% of all dwellings has been achieved in the study area. In Kilinochchi 8 sub-areas were selected by the AMC on the basis of:

- the population size of an sub-area, such that the required sample size could be obtained,
 - the migration habits of the population in the sub-area; viz. a relatively static population with minimum migration was required,
 - the homogeneity of the population; all children less than 6 years living in a sub-area had to be exposed to the same risk of receiving an inoculum.
- The condition of homogeneity of a sub-area has been judged by the AMC. About 1/4 of the Kilinochchi area is covered by the 8 sub-areas, which are identified and coded in the table below. A total of 31 villages were included in

the survey.

Sub-area identification

code	sub-area	code	sub-area
1	Chempianpattu	5	Poonakary
2	Pallai	6	Kariyalainagapaduwan
3	Iyakkachchi (first village name)	7	Mallavi
4	Uruthirapuram	8	Akkarayankulam

To obtain the necessary data it was planned to have for each sub-area at least 50 children examined in each of the following age groups: 0.5-1.5yr, 1.5-2.5yr, 2.5-3.5yr, 3.5-4.5yr, 4.5-5.5yr. Hence the aim was to have at least 250 samples in each sub-area and at least 2000 children under the age of 6 years examined in the survey. This survey was conducted in late September-October 1978. One sub-area was sampled each day. If a sub-area was too wide spread the sampling was spread over a period of 2 days. Advance propaganda was carried out on the day previous to collection and the mothers were asked to bring the children to the collecting centres at appointed times.

Blood from a fingerprick was collected in 2 heparinized capillaries and a thick film made for parasitological examination. The capillaries with blood were centrifuged in the field and the sera stored on ice till they were brought to Colombo. In Colombo they were stored at -20°C until the sera were processed. The sera were analysed using the IFA test, adapted from the method as described by Voller and O'Neill (1971), with *P. fieldi* as antigen. Fluorescein conjugated anti-human globulin (Nordie) was used at a concentration of 1/200. A titre of 1/40 was considered positive. A Leitz Ortholux incident light fluorescence microscope with a Xenon 75 watt illumination was used.

The serological data were analysed using the spatial-dependent and time-

dependent transmission model; the statistical inference was based on the F-design respectively R-design approach (see section 3.3.3). The obtained average trend in transmission intensity in the 8 sub-areas (F-design) and the area Kilinochchi (R-design) is compared with the moving annual trend derived from quarterly parasitological figures. The parasitological data were obtained from an independent retrospective investigation carried out by the AMC in the period July to September 1979. For each sub-area quarterly parasitological figures for the years 1973 to 1978 were collected (total blood smears examined 14,943).

3.4.3 Analysis of serological data

Table 3.2 (see page 79) presents the age related seropositivity rates of the children examined in each sub-area. The total number of children serologically examined in September-October 1978 was 1,678 with an overall seropositivity rate of 16% (IFA test, *P. fieldi*, pos=titre \geq 1:40). The overall test for the detection of spatial variation in the serological profiles of the 8 subpopulations is very significant (Chi-square test, see section 3.3.1, degrees of freedom 35, $p=10^{-5}$).

Analysis on sub-area level

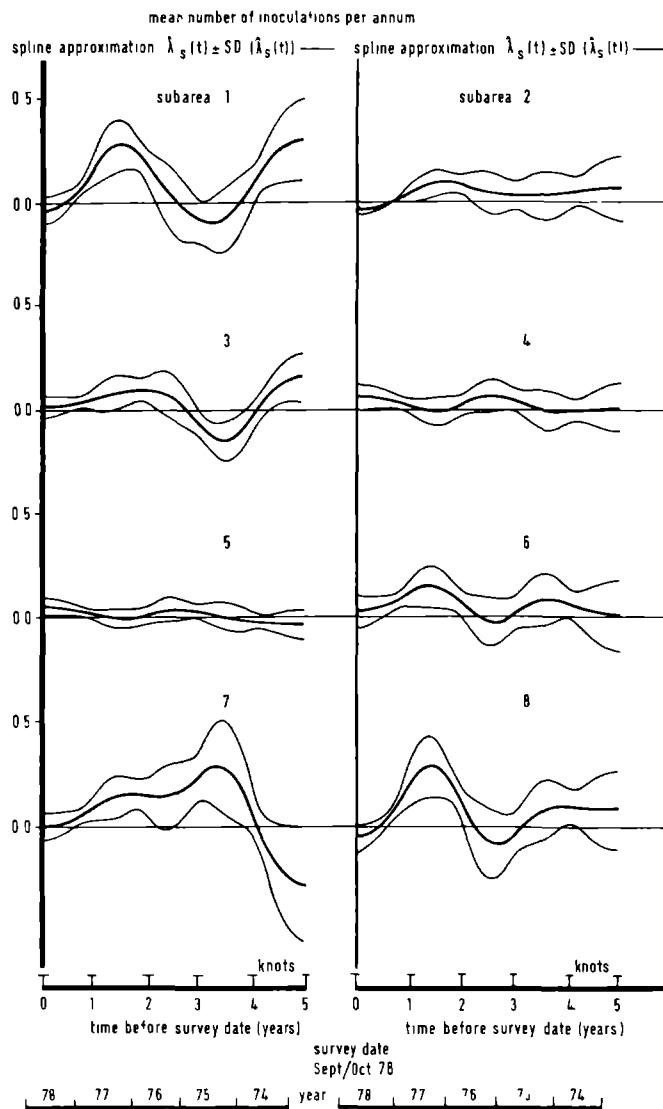
The time-dependent transmission model, described in chapter 2, has been used to assess the trend of transmission in each sub-area. The spline approximations $\hat{\lambda}_s(t) \pm SD(\hat{\lambda}_s(t))$ for each sub-area are shown in fig. 3.2 (see page 80). The knots of the spline approximation are situated at the time points 1, 2, 3, 4, 5 years before survey date.

To detect changes in the annual trend of the transmission intensity in each sub-area the two-sided test described in chapter 2 has been used.

Table 3.2 Results of IFA test carried out on serum samples collected in 8 sub-areas of the Kilinochchi district, Sri Lanka, in 1978 using *P. fieldi* as the antigen (positive is titre $\geq 1:40$).

age group (yr)	no exam.			no pos.			% pos.			no exam.			no pos.			% pos.					
	sub-area 1						sub-area 2						sub-area 3						sub-area 4		
0.5-1.5	30	1	3	30	0	0	30	0	0	38	2	5	32	1	3	38	2	5			
1.5-2.5	45	11	24	44	4	9	44	5	11	50	3	6	44	7	16	51	6	12			
2.5-3.5	47	12	26	42	6	14	37	2	5	48	6	13	45	8	18	47	11	23			
3.5-4.5	37	8	22	45	8	18	45	5	11	46	6	13	45	19	38	47	11	23			
4.5-5.5	50	19	38	47	11	23	32	5	15	38	2	5	34	5	15	46	6	13			
	sub-area 5						sub-area 6						sub-area 7						sub-area 8		
0.5-1.5	35	1	3	35	2	6	43	1	2	52	9	17	35	1	3	37	1	3	35	1	3
1.5-2.5	43	1	2	52	9	17	38	2	5	57	10	18	43	7	16	29	7	24	44	13	30
2.5-3.5	38	2	5	47	11	23	37	2	5	47	11	23	44	15	35	46	10	22	43	17	39
3.5-4.5	37	2	5	47	11	23	36	1	3	47	12	26	43	15	35	46	15	33	44	19	45
4.5-5.5	36	1	3	47	12	26	35			37			43			46			44		

Figure 3.2 The annual spline-trend in transmission intensity $\hat{\lambda}_s(t) \pm SD(\hat{\lambda}_s(t))$ in eight sub-areas of the Kilinochchi district, Sri Lanka.



Periods before survey date in which the two-sided p-value function takes on values less than 0.05 are considered to indicate a change in the trend of transmission. The results are presented in table 3.3. No significant changes in the trend of transmission could be established in the sub-areas 4, 5, 6 and 7. The minimum values of the two-sided p-value functions for these areas were respectively 0.52, 0.38, 0.38 and 0.14 respectively. Sub-area no. 1, 2 and 8 show evidence of a decrease in the annual trend of the transmission intensity about 1.3 years before survey date ($p \leq 0.05$). A significant increase is found about 2.5 years before survey date in sub-area no. 1 and sub-area no. 3.

Table 3.3 Application of the time-dependent transmission model for eight sub-areas of the Kilinochchi district, Sri Lanka; change in spline trend $\lambda_s(t)$, two-sided test.

Trend-change test			Time before survey date (years)	
sub-area no	minimal p-value	change in trend	decrease of $\hat{\lambda}_s(t)$, $p \leq 0.05$	increase of $\hat{\lambda}_s(t)$, $p \leq 0.05$
1	0.03	yes	1.3	2.4-2.5
2	0.01	yes	1.2-1.3	
3	0.04	yes		2.5
4	0.52	no		
5	0.38	no		
6	0.38	no		
7	0.14	no		
8	0.04	yes	1.3	

Fig. 3.3 provides in various subfigures information on the average trend of transmission in the eight combined sub-areas (fixed design) and the Kilinochchi district (random design). The estimates $\hat{\lambda}_s(t)$ of the spline-trends in transmission intensity in the 8 sub-areas are shown in subfigure A of figure 3.3. The unweighted mean $\bar{\lambda}_s(t)$ of these eight functions is presented separately in subfigure B. This average trend function $\hat{\lambda}_s(t)$ is considered a) an estimate of the average annual trend in transmission in the eight combined sub-areas (F-design) and b) an estimate of the average annual trend in transmission in the Kilinochchi district (R-design). The trend function $\hat{\lambda}_s(t) \pm$ standard error $SD(\hat{\lambda}_s(t))$ for these two designs are presented in the subfigures C and E. Evidence of change in trend of transmission in the eight combined sub-areas and the Kilinochchi district is stored in the two-sided p-value functions $P_F(t)$ and $P_R(t)$ respectively (subfigures D and F). Periods in which the p-value function takes on values less than 0.05 are considered to reflect a change in the average trend of transmission. According to this convention the following results are obtained:

fixed design

In period 0.0-1.3 yr before survey date the average trend in transmission intensity in the 8 sub-areas together is decreasing, whilst an increasing trend is found 2.4 years before survey date.

random design

In period 0.0-1.4 yr before survey date the average trend in transmission intensity in Kilinochchi is decreasing, about 2.3 years before survey date there is an indication of an increasing trend.

Figure 3.3 The average trend-function $\hat{\lambda}_s(t)$ as an estimate of the average trend in transmission in eight combined sub-areas (fixed design) and the Kilinochchi district (random design); detection of change in the trend of transmission (two-sided test).

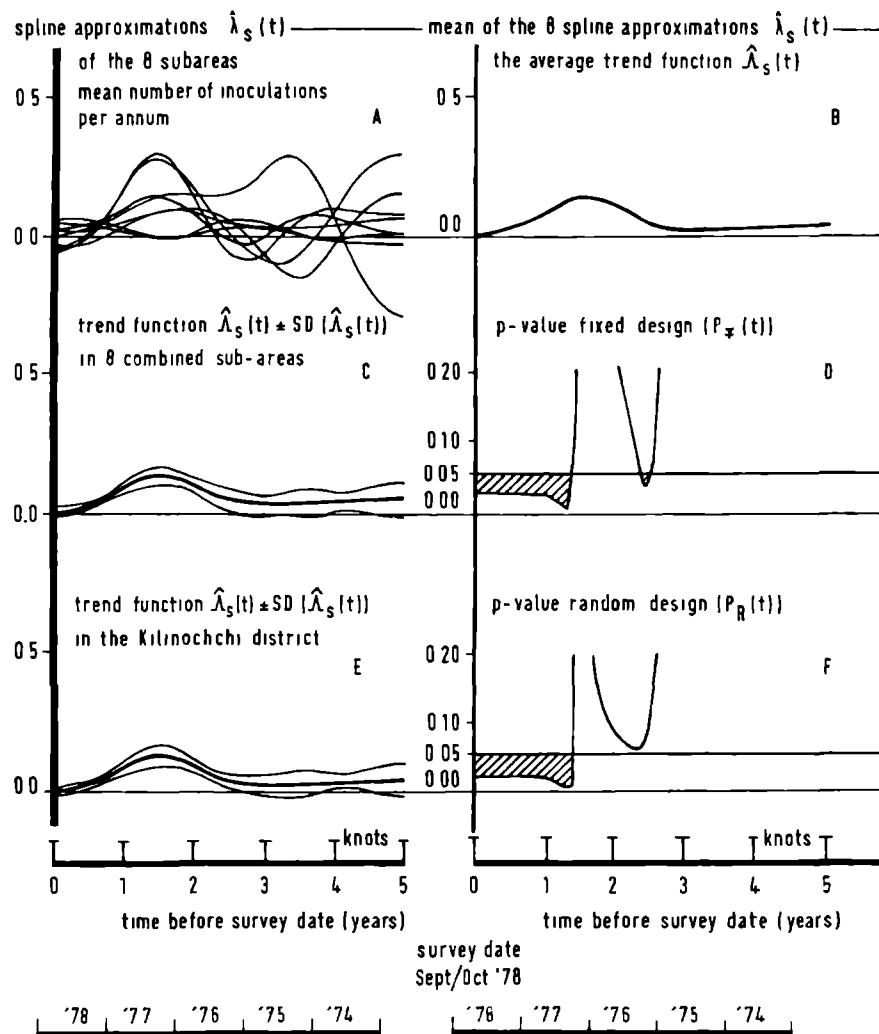


Fig. 3.4 shows - for comparison purposes - the one-sided p-value function $P_F(t)$ of the fixed design, the one-sided p-value function of the test based on the sum of the standardized rates of change of $\hat{\lambda}_s(t)$ in the sub-areas and the one-sided p-value function based on the combination of the 8 one-sided p-value functions associated with the separate spline approximations $\hat{\lambda}_s(t)$. The periods before survey date in which these one-sided p-value functions take on values less than the critical value 0.05 are:

test based on	Periods before survey date (yr)	
	decrease	increase
fixed design	0.0-1.4	2.3-2.5
$\sum \hat{\gamma}_s(t) / SD(\hat{\gamma}_s(t))$	0.0-1.3	2.4-2.5
combination p-values	0.0-1.3	2.4-2.5

Spatial variation

Fig. 3.5 (see page 86) presents for each time point t before survey date the square root of the total variance and of the spatial variance with respect to the variation in $\hat{\lambda}_s(t)$ and $\hat{\gamma}_s(t)$ respectively (F-design). In this figure is also shown the proportion explained variance of the total variance in $\hat{\lambda}_s(t)$ and $\hat{\gamma}_s(t)$ respectively. These proportions¹⁾ are indicated by $E_\lambda(t)$ and $E_\gamma(t)$.

Pairwise comparison of sub-areas

Table 3.4 (see page 87) presents the results of the pairwise comparison of the 8 sub-areas using the overall test for comparing two homogeneous populations (see section 3.2). Those pairs of sub-areas where there were

¹⁾ The values of $E_\lambda(t) \geq 1$ and $E_\gamma(t) \geq 1$ are not represented in fig. 3.5.

Figure 3.4 Three methods to combine evidence of change in the trend of transmission. Illustration using serological data from eight sub-areas of the Kilinochchi district; p-value functions (one-sided).

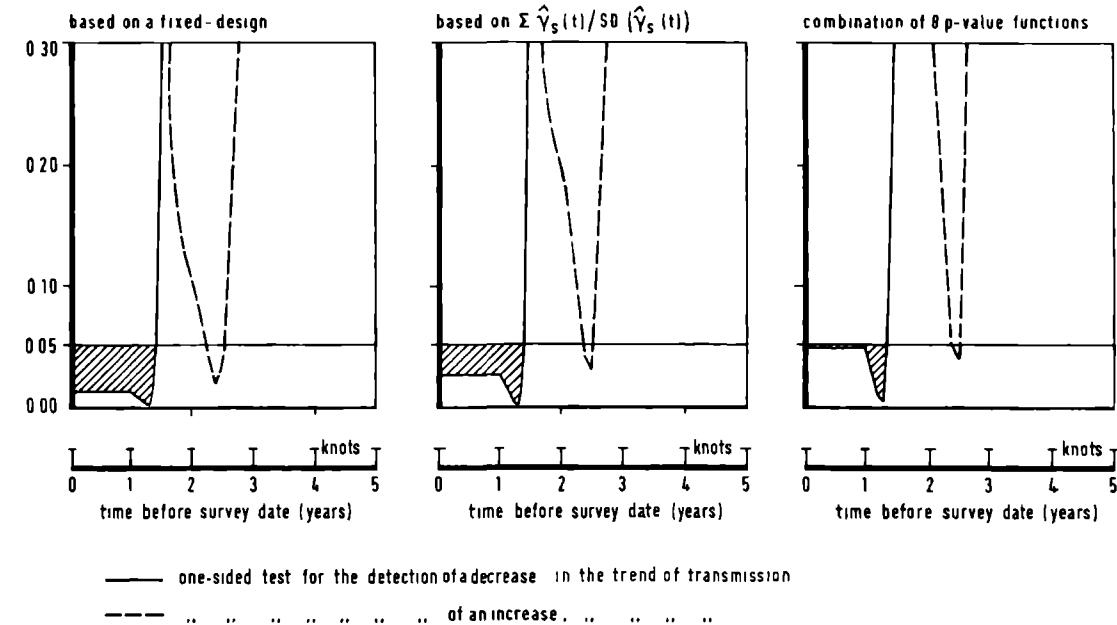


Figure 3.5 Spatial variation in the trend of transmission in eight sub-areas of the Kilinochchi district (fixed design).

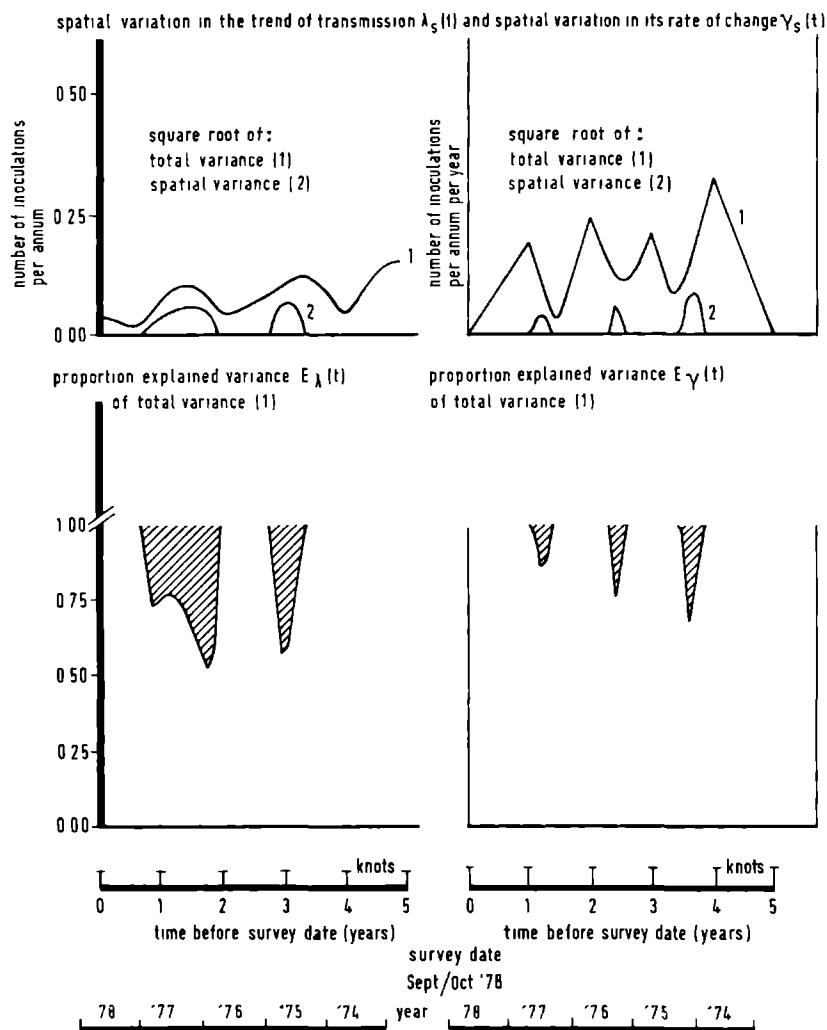


Table 3.4 Pairwise comparison of eight sub-areas of the Kilinochchi district, Sri Lanka. Overall Chi-square test for comparing two homogeneous populations.

no sub-area	Chi-square test, p-values							
	1	2	3	4	5	6	7	8
1	0.34	0.07	0.01	0.00	0.83	0.47	0.99	
2		0.80	0.91	0.11	0.93	0.10	0.61	
3			0.97	0.37	0.42	0.00	0.09	
4				0.74	0.36	0.00	0.05	
5					0.01	0.00	0.00	
6						0.37	0.99	
7							0.62	
8								

indications for a difference in trend of transmission intensity ($p \leq 0.05$) were investigated further using the "trend-value test" and "trend-change test". The results are presented in table 3.5 (see page 88). Differences in the rate of change in intensity occur in the periods 1.2-1.3 yr and 2.4-2.5 yr respectively before survey date (trend-change test $p \leq 0.05$).

Parasitological examination

The thick blood films of all 1,678 samples were examined at the Anti Malaria Campaign. Three¹⁾ were positive for Plasmodium vivax (sub-area 1, 3 and 8).

¹⁾These three children were also serologically positive

Table 3.5 Pairwise comparison of eight sub-areas of the Kilinochchi district, Sri Lanka.
 Further investigation using the trend-value test and the trend-change test for two homogeneous populations.

Overall Chi-square test for comparing two homo- geneous populations			Trend-value test periods before survey date (yr) with different trend values $\lambda_s(t)$			Trend-change test periods before survey date (yr) with different rates of change in $\lambda_s(t)$		
no sub-area			no sub-area			no sub-area		
3	4	5	3	4	5	3	4	5
sub-area 1	*	**		1.3-1.9	0.8-1.9		2.4	1.2-1.3 2.4-2.5
sub-area 6		**			0.9			
sub-area 7	**	**	**	2.9-3.3		1.8		
sub-area 8	*	**		1.6-1.8	1.3-1.9			

* = $0.01 < p \leq 0.05$

** = $p \leq 0.01$

differences are found in the
periods ($p \leq 0.05$):

0.8-1.9

2.9-3.3

differences are found in the
periods ($p \leq 0.05$)

1.2-1.3

2.4-2.5

3.4.4 Analysis of parasitological data for the years 1973-1978

The conventional parasitological parameters for the assessment of the epidemiological trend are the number of laboratory confirmed malaria cases per year, the annual parasite incidence (API) and the slide positivity rates (SPR) observed in repeated parasitological surveys¹⁾. The laboratory confirmed malaria cases for the years 1973 to 1978 could be collected only by going through blood film records for the last 6 years, because neither those who carried out the surveillance operations²⁾ nor the laboratory or even the regional and subregional offices of the Anti Malaria Campaign (AMC) had kept records of malaria incidence statistics for individual villages. Unfortunately the laboratory where the blood films were examined did not file these records. As such the statistics were obtained from whatever copies of the blood film records that were available. Furthermore the surveillance operations were

¹⁾ The annual parasite incidence (API) is the number (per thousand of a population) of laboratory confirmed malaria cases detected during one year. The slide positivity rate (SPR) is the number of positive slides in relation to the number of blood smears examined during a given time period.

²⁾ One of the activities of surveillance operations is concerned with malaria case detection. A malaria case is the occurrence of a malaria infection in a person in whom, regardless of the presence or absence of clinical symptoms, the presence of malaria parasites in the blood has been confirmed by microscopic examination. The purpose of case detection is to identify malaria-infected persons, treat them and furnish the necessary data for assessment of the malaria situation. Case detection is a screening process, using as its indicator either the symptom of fever or specific epidemiological attributes. There are two types of case detection, active and passive. Active case detection is the process of case-finding by visiting, at regular intervals, all houses of a malarious area and taking blood specimens of any inhabitants who have, or have recently had, fever. Passive case detection is the finding of malaria cases through notification by medical or other collaborating personnel to whom fever cases and other suspected cases are reported. Passive case detection is mainly done by malaria detection posts (hospitals, clinics, medical practitioners and voluntary collaborators).

only implemented intermittently at medical institutions. Hence active and passive surveillance coverage was incomplete in space and time. The population sizes of the sub-areas were not available, therefore the API could not be calculated. Although the figures collected do not fulfil the criteria for admission of survey data for epidemiological evaluation, the parasitological figures have been used to assess the trend of the SPR in the eight sub-areas.

The total number of blood smears examined during a period of 5 years before Oct. 1978 in the 8 sub-areas was 14,943. The number of laboratory confirmed malaria cases was 3,237, resulting in an overall slide positivity rate (SPR) of 22%. Table 3.6 (see page 91) presents the annual number of blood smears examined, the number of positive cases and the slide positivity rate (SPR) by sub-area and by year.

The "moving annual trend" has been chosen as a measure of the trend in the quarterly slide positivity rates in a sub-area. Let q_{i-2} , q_{i-1} , q_i , q_{i+1} , q_{i+2} be the quarterly slide positivity rates in the quarters $i-2$, $i-1$, i , $i+1$, $i+2$. The moving annual SPR at quarter i ($m(i)$) is defined by¹⁾

$$m(i) = \frac{1}{2}(m_1(i) + m_2(i)), \quad (3.46)$$

where

$$m_1(i) = \frac{1}{4}(q_{i-2} + q_{i-1} + q_i + q_{i+1})$$

$$m_2(i) = \frac{1}{4}(q_{i-1} + q_i + q_{i+1} + q_{i+2}).$$

The terms $m_1(i)$ and $m_2(i)$ are means of four adjacent quarterly SPR rates. Using $m_1(i)$ and $m_2(i)$ the seasonal fluctuations in the quarterly SPR rates are eliminated. In order to centre the moving annual trend value in quarter i , $m(i)$ is defined as the mean of $m_1(i)$ and $m_2(i)$. The value of $m(i)$ in quarter i

¹⁾ The sequence of quarters $i-2$, $i-1$, i , $i+1$, $i+2$ refers to the conventional direction of the time-scale, i.e. quarter $i+2$ is one year after quarter $i-2$.

Table 3.6 Laboratory confirmed malaria cases, number of blood smears examined and slide positivity rate (SPR) during a period of 5 years before October 1, 1978 in 8 sub-areas of the Kilinochchi district, Sri Lanka.

year before survey date	no exam.	no pos.	SPR (%)	no exam.	no pos.	SPR (%)
sub-area 1				sub-area 2		
1 th	104	1	1	305	31	10
2 th	295	51	17	376	80	21
3 th	283	63	22	217	21	10
4 th	475	125	26	222	24	11
5 th	1076	338	31	96	6	6
sub-area 3				sub-area 4		
1 th	185	12	6	25	1	4
2 th	349	74	21	709	92	13
3 th	527	93	18	596	75	13
4 th	315	60	19	471	104	22
5 th	327	102	31	349	115	33
sub-area 5				sub-area 6		
1 th	54	3	6	107	9	8
2 th	534	105	20	388	86	22
3 th	229	38	17	457	81	18
4 th	173	34	20	337	75	22
5 th	219	33	15	267	81	30
sub-area 7				sub-area 8		
1 th	288	36	13	50	2	4
2 th	564	112	20	138	25	18
3 th	678	137	20	330	89	27
4 th	1207	336	28	622	245	39
5 th	787	204	26	212	28	13

depends on the values of the quarterly slide positivity rates in the quarters $i-2$, $i-1$, i , $i+1$, $i+2$ (see (3.46)). In particular $m(i)$ is influenced by $q(i+2)$. Therefore a relatively low slide positivity rate $q(i+2)$ in quarter $i+2$ induces a relatively low trend value $m(i)$ two quarters before the quarter $i+2$.

Fig. 3.6 (see page 93) shows the quarterly figures of the SPR rate by sub-area in the last five years before Oct. 1978 (the survey date of the cross-sectional serological survey) and the moving annual trend derived from these quarterly figures. In the years 1977 and 1978 quarterly parasitological figures were not always available (or the number examined was less than five) in the following sub-areas: sub-area no. 1 (1977, 4th), sub-area no. 4 (1977, 4th; 1978, 1st and 2nd), sub-area no. 5 (1977, 4th; 1978, 1st), sub-area no. 6 (1977, 1st and 4th; 1978, 1st), sub-area no. 7 (1977, 4th) and sub-area no. 8 (1978, 1st and 2nd). These "missing values" are given in fig. 3.6 by linear interpolation using the adjacent known quarterly slide positivity rate figures.

Of special interest is the course of $m(i)$. It is readily verified that the difference between two adjacent values of $m(i)$ is given by (see (3.46))

$$m(i+1) - m(i) = \frac{1}{8}(q_{i+2} + q_{i+3} - q_{i-2} - q_{i-1}). \quad (3.47)$$

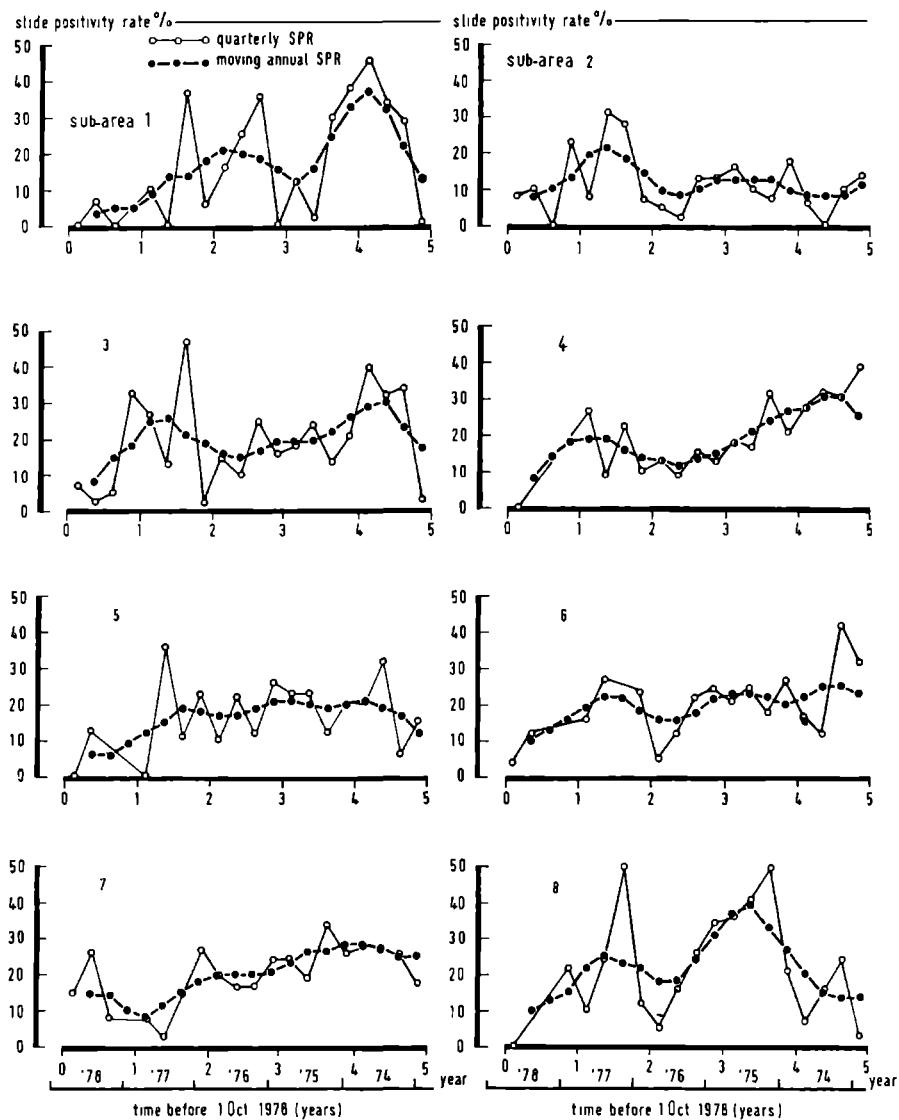
Hence a change in the moving annual SPR trend in the quarters i to $i+1$ is determined by the quarterly slide positivity rates in the quarters $i-2$, $i-1$ and $i+2$, $i+3$. To detect a change in the moving annual SPR trend in quarter i to $i+1$ the slide positivity rates in the adjacent quarters i.e. q_{i-2} , q_{i-1} , q_{i+2} , q_{i+3} are used. Separately for each sub-area and each i the following hypothesis

$$H_0: q_{i-2} = q_{i+2} \text{ and } q_{i-1} = q_{i+3}$$

is tested against the two-sided alternative

$$H_1: \text{generally the differences } q_{i+2} - q_{i-2}, \\ q_{i+3} - q_{i-1} \text{ have the same sign.}$$

Figure 3.6 The quarterly slide positivity rates (SPR) and the moving annual trend in 8 sub-areas of the Kilinochchi district in a period of 5 years before 1. October 1978.



The test-statistic used is

$$t_i = (q_{i+2} - q_{i-2}) + (q_{i+3} - q_{i-1}). \quad (3.48)$$

Under H_0 the variable t_i is approximately normally distributed with mean 0 and variance c_i given by (Rümke and van Ueden (1961); combination of two (2x2) contingency tables).

$$c_i = \frac{a_i(n_{i+2} + n_{i-2} - a_i)}{(n_{i+2} n_{i-2})(n_{i+2} + n_{i-2} - 1)} + \frac{b_i(n_{i+3} + n_{i-1} - b_i)}{(n_{i+3} n_{i-1})(n_{i+3} + n_{i-1} - 1)} \quad (3.49)$$

where

n_i = number of slides examined at quarter i

$$a_i = q_{i+2} n_{i+2} + q_{i-2} n_{i-2}$$

$$b_i = q_{i+3} n_{i+3} + q_{i-1} n_{i-1}.$$

Using $t_i/\sqrt{c_i}$ a two-sided p-value has been calculated to detect a change in the moving annual SPR trend at quarter i to $i+1$; a p-value ≤ 0.05 is considered as an indication for a change in $m(i)$. Table 3.7 (see page 95) presents the results for each sub-area.

To detect a change in the overall annual SPR trend in the eight sub-areas the results of the statistical tests performed in all individual sub-areas have to be combined. Let $q_{i,j}$ be the quarterly slide positivity rate at quarter i in sub-area j ($j=1, \dots, 8$). Furthermore let t_{ij} and c_{ij} be the statistics (3.48) and (3.49) in sub-area j ($j=1, \dots, 8$). To test the hypothesis

$$H_0: q_{i-2,j} = q_{i+2,j} \text{ and } q_{i-1,j} = q_{i+3,j} \quad (j=1, \dots, 8)$$

against the two-sided alternative

H_1 : generally the differences $q_{i+2,j} - q_{i-2,j}$, $q_{i+3,j} - q_{i-1,j}$ ($j=1, \dots, 8$) have the same sign,

the sum of the t_{ij} ($j=1, \dots, 8$) is used,

$$T_i = \sum_{j=1}^8 t_{ij}. \quad (3.50)$$

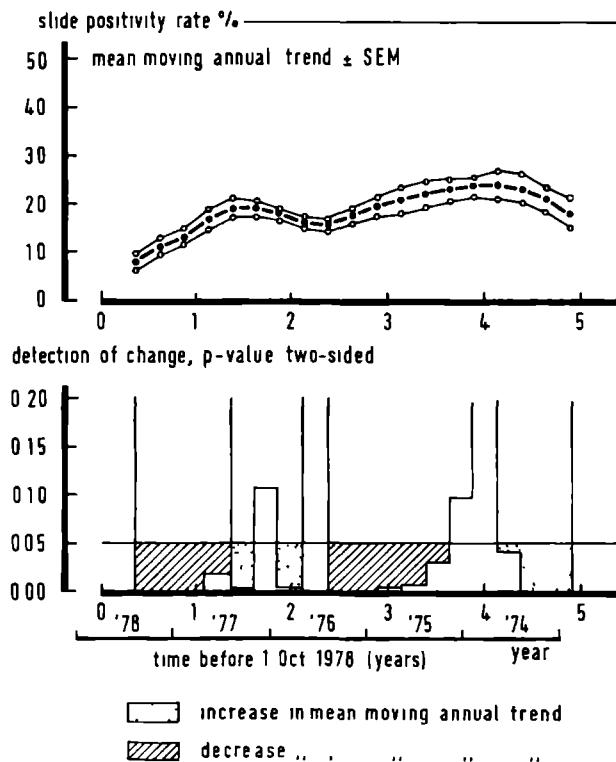
Table 3.7 Detection of change in the moving annual slide positivity rate (SPR) for each sub-area and in the eight combined sub-areas of the Kilinochchi district.

year	(years)	Change in the moving annual SPR trend ($p \leq 0.05$)								overall	
		+ = increase									
		no. sub-area									
time before 1 Oct. 1978		1	2	3	4	5	6	7	8		
74.0	4.75	+	-	+	+	+				+	
.25	4.50	+		+						+	
.50	4.25	+					-	+	+	+	
.75	4.0	-		-					+		
75.0	3.75	-	+	-	-				+		
.25	3.50	-		-	-			+		-	
.50	3.25	-		-			-			-	
.75	3.0	+		-			-	-		-	
76.0	2.75	+	-	-	-		-			-	
.25	2.50		-	-			-			-	
.50	2.25										
.75	2.0		+	+			-			+	
77.0	1.75	-	+				+	-			
.25	1.50		+	+	+		+	-		+	
.50	1.25		-							-	
.75	1.0	-	-	-		-		+		-	
78.0	0.75	-	-	-	-	-	-	+	-	-	
.25	0.50	-	-	-	-		-			-	

Under the null hypothesis T_i is approximately normally distributed with mean 8 and variance $C_i = \sum_{j=1}^8 c_{ij}$. The corresponding p-value has been calculated to detect in quarter i to $i+1$ a change in the moving annual SPR trend in the 8 combined sub-areas. The results are also presented in table 3.7.

The mean of the 8 moving annual SPR trends in the sub-areas with the standard error of the mean (SEM) is shown in fig. 3.7 (see page 96); the p-values of the overall test (test statistic (3.50)) are presented in the

Figure 3.7 Detection of change in the mean moving annual trend of the slide positivity rate (SPR) in eight combined sub-areas of the Kilinochchi district, Sri Lanka.



lower-half of the figure. Adjacent quarters for which the p-value assumes a value less than 0.05 are considered to indicate a change in the mean moving annual SPR trend.

3.5 DISCUSSION

Before the results of the application of the transmission model in Kilinochchi are discussed and compared with the parasitological data, a few remarks regarding the methodology are made.

3.5.1 Remarks regarding methodology

Spatial-dependent and time-dependent transmission model

The model described in this chapter is basically a time-dependent transmission model with a stratification to one factor or covariate (here the factor area). The non-homogeneous population is divided into subpopulations or strata. It is easily understood that strata may be defined in terms of more than one variable. For example strata may be defined using characteristics such as ethnic group, climatic condition or type of malaria control. The important point is that within strata the variability in the risk of receiving inoculations should be as low as possible.

In analysing the serological data from Sri Lanka the parameter $\lambda_s(t) = \sum w_i \lambda_{i,s}(t)$ was used with equal weightage w_i as a measure of the average trend in transmission intensity. An equivalent approach is to use the concept average cumulative inoculation rate $P(A)$ and analyse this rate $P(A)$ - with appropriate adaptation of sampling errors - using the time-dependent transmission model. It should be emphasized that $1-P(A)$ is the (weighted) geometric mean of $1-p_i(A)$.

As has been demonstrated in section 3.3.2, misleading results might be obtained by the transmission model which uses the crude cumulative inoculation rate $p(A)$ of the non-homogeneous population (see fig. 3.1^{a, b}). With respect to the Kilinochchi data, however, these possible artefacts do not appear.

Application of the time-dependent transmission model to the age related

seropositivity rates of all sub-areas together provides the same results as the results obtained by application of the spatial-dependent and time-dependent transmission model (fixed design). This is explained by the almost equal figures of the crude seropositivity rates $p(A)$ and the average seropositivity rates $P(A)$ (see table below).

age related seropositivity rates (%)					
	age group (yr)				
	0.5-1.5	1.5-2.5	2.5-3.5	3.5-4.5	4.5-5.5
crude rate $p(A)$	3.4	13.4	17.8	19.9	24.1
average rate $P(A)$	3.3	14.2	18.0	20.7	23.9

As regards the selection of sub-areas it cannot be claimed that the 8 sub-areas included in the survey are a (weighted) random sample of sub-areas from Kilinochchi (section 3.4.2). Furthermore the population sizes of the subpopulations living in these sub-areas were not available. It was therefore decided to perform an unweighted analysis based on a fixed design and random design approach. The analysis of the data using the R-design is based on the questionable assumption that the sample of 8 sub-areas can be regarded as effectively random.

As discussed in chapter 2, the position of the knots determines the type of trend that is assessed. In this application the distance between the knots is one year. Hence the spline approximation $\Lambda_s(t)$ describes the (unweighted) average annual trend of transmission. The property of preservation of surface and the minimum change property of $\Lambda_s(t)$ explain why $\Lambda_s(t)$ has been chosen as a measure of the trend of transmission intensity in a non-homogeneous population.

In comparing the trend $\Lambda_s(t)$ based on serological data with the trend based on the slide positivity rate, the SPR trend, it should be clear that both trends are reflecting different aspects of the same phenomenon. Therefore it cannot be expected that, apart from statistical fluctuations, both approaches will provide exactly the same results. The serological evidence of a change in the trend of the transmission intensity is based on the sero-reactivity in children of age 0.5-5.5 years in September-October 1978. However, the SPR trend is based on the examination of blood smears of individuals of all ages in the years 1973-1978. The SPR trend reflects the trend of the laboratory confirmed malaria cases in relation to the total number of blood smears examined, it is a moving annual trend of quarterly figures. The spline approximation $\Lambda_s(t)$, however, intends to measure the annual trend of the inoculation intensity. So for the following main reasons it cannot be expected that both methods will produce identical results:

- an inoculation does not necessarily result in a parasitaemia
- the annual spline-trend differs from the moving annual trend of quarterly figures.
- both methods possess their own sources of bias (to be discussed in the next two subsections).

Biased results in the analysis of serological data

It is emphasized that application of the spatial-time dependent transmission model to serological malaria data definitely will cause biased results since the model does not take into account a) the age related heterogeneity in the risk of receiving inoculations and b) the factor fading of antibodies.

The assumption of equal risk is only required for the individuals of the age band included in the survey and not for the total subpopulation. In the survey in Kilinochchi children were examined in the age class 0.5-5.5 yr.

It is therefore sufficient to assume equal risk of receiving an inoculation for all children in a sub-area under the age of 6 years at a time point t years before survey date. As has been pointed out in chapter 2 (section 2.4.3), the assumption of homogeneity even in a small village and a small age band remains doubtful.

With respect to b) it is assumed that the survey provides the means to separate those who have already experienced an infection from those who have not. This is the most questionable assumption; the IFA test (*P. fiedli* antigen, pos=titre $\geq 1:40$) is certainly not capable of making this distinction. To assess the specificity of the IFA test 88 children from a non malarious area were serologically examined. The distribution of these children over the age groups 0.5-2.0 yr, 2.0-3.0 yr, 3.0-4.0 yr, 4.0-5.0 yr, 5.0-6.0 yr was respectively 20, 17, 19, 12, 20. All these children were negative at a dilution of 1:40, indicating a specificity close to 1. Therefore although the proportion false positives amongst the positives (titre $\geq 1:40$) is probably low, the proportion false negatives amongst the negatives (titre $< 1:40$) may be considerable. In chapter 4 a correction factor is introduced to compensate for the bias in $\hat{\lambda}_s(t)$ and $\hat{\Lambda}_s(t)$ caused by the fading of antibodies.

Biased results in the analysis of parasitological data

With respect to the reliability of the parasitological data the following additional remarks have to be made. No figures of the annual parasite incidence (API) in the sub-areas were available. Only figures of the laboratory confirmed malaria cases and the slide positivity rate (SPR) were collected. Moreover it cannot be claimed that the quarterly figures of the slide positivity rate in fig. 3.6 represent a constant proportion of cases that have occurred for the following reasons:

- In sub-area 2, 5 and 8 not all blood film records were available
- The passive surveillance agency which contributed much more blood smears than the active surveillance agent, did blood filming only intermittently in 1976 and early 1977 due to lack of glass slides. Further blood filming was completely stopped from September 1977 following the revision of the surveillance strategy in the AMC. The figures furnished for 1978 are mostly based on active surveillance and activated passive surveillance¹⁾.
- Active surveillance records for 1973 were not available for sub-areas 2-7, the cases recorded have been based entirely on passive surveillance blood filming. Furthermore active surveillance had not been carried out from August 1977 to March 1978 in these sub-areas. Two of the three villages in sub-area no. 5 had not been under active surveillance until the study started in 1978.

Further confusion did arise as not all the blood film records traced did have laboratory results recorded on it. These records were rejected. It will be clear from the above that the annual trend of the number of laboratory confirmed malaria cases is not a reliable basis for an evaluation of the trend in transmission. In the hope that the annual SPR trend still may reflect some reality the quarterly SPR figures have been analysed.

With the acknowledgement of the shortcomings of the analysis of the serological data as well as the analysis of the parasitological data a comparison will be made between $\hat{\Lambda}_s(t)$ and the mean moving annual SPR trend.

¹⁾ Activated passive surveillance refers to the situation in which a passive case detection agency is permanently manned by a staff-member of the Anti Malaria Campaign (AMC).

3.5.2 Comparison of the spline-trend $\hat{\lambda}_s(t)$ with the mean SPR trend

The change in the mean annual SPR trend (fig. 3.7) supports the general conclusions drawn from the serological data using the spatial-time dependent transmission model (fig. 3.3). Both methods indicate an increasing trend of malaria transmission which starts in the middle of 1976 and both methods indicate a downward trend after the middle of 1977 (fig. 3.3, fig. 3.7). The results of the analysis of the cross-sectional serological data of the 1978 survey and those of the parasitological data for the years 1976-1978 apparently reflect reality or the results using both methods are artefacts. The sources of such probable artefacts are completely different in the two methods. The main factor which may produce a false result in the course of $\hat{\lambda}_s(t)$ appears to be the occurrence of the fading of antibodies whereas the changing policies of the national, regional and subregional institutions of the Anti Malaria Campaign are responsible for errors in the course of the SPR trend. Therefore it might well be that the application of the spatial-time dependent transmission model did not produce false results with regard to the detection of periods of change in the trend of transmission. The indicated change in the SPR trend in the years 1974-1975, however, is not reflected in the course of the spline-trend $\hat{\lambda}_s(t)$.

3.5.3 Comparison of the spline-trend $\hat{\lambda}_s(t)$ with the SPR trend at sub-area level

The results of the application of the time-dependent transmission model presented in table 3.3 are not inconsistent with the course of the SPR trend in the corresponding sub-areas (fig. 3.6 and table 3.7). The p-value functions for the detection of periods of change in $\lambda_s(t)$ in the sub-area no. 4, 5, 6 and 7 were not "significant". Therefore the course of $\hat{\lambda}_s(t)$ in these sub-areas do not allow conclusions with regard to a changing trend. This is as

such not contradictory to the course of the SPR trend in these sub-areas.

Larger sample sizes in the sub-areas might result in statistically significant changes in the sub-areas 4, 5, 6 and 7.

The course of $\hat{\lambda}_s(t)$ in sub-area no 3 is to a large extent the result of the relatively low seropositivity rate (5%) in the age group 3.5-4.5 yr in this sub-area (table 3.2). This relatively low percentage might be the result of just chance.

Generally the levels of $\hat{\lambda}_s(t)$ do not fit well with the levels of the SPR trend. The curves $\hat{\lambda}_s(t)$ in fig. 3.2 are very likely quite biased. Therefore the results of the trend-value test and trend-change test for the comparison of two homogeneous populations are probably not reliable; the major cause being the neglect of the fading of antibodies. Differences in the rate of change of $\hat{\lambda}_s(t)$ between two sub-areas are established in the periods 1.2-1.3 yr and 2.4-2.5 yr before survey date (table 3.5). In period 1.2-1.3 yr before survey date $\hat{\lambda}_s(t)$ shows a decreasing trend in sub-area no 1 while in sub-area no 5 such a decreasing tendency is not found (fig. 3.2). The SPR trends in sub-area no 1 and no 5, however, both show a decreasing tendency in period 1.2-1.3 yr before survey date (fig. 3.6). About 2.4 years before survey date an increase in $\hat{\lambda}_s(t)$ is observed in sub-area no 1 while sub-area no 4 and sub area no 5 do not show such an increase in the same period (fig. 3.2). To some extent this difference between the sub-areas no 1, 4 and 5 is reflected by the trend in the SPR data in the corresponding period (fig. 3.6).

The overall impression is that although $\hat{\lambda}_s(t)$ and $\hat{\Lambda}_s(t)$ are biased, "statistically significant" changes in these functions still may reflect true changes in the trend of transmission.

3.5.4 Indications for the effectiveness of malathion

In September-October 1977, exactly one year before the survey date of the 1978 cross-sectional survey, DDT has been replaced by malathion in the area of Kilinochchi. For the detection of a change in the average trend of transmission in the eight combined sub-areas, the p-value function of the F-design has been used (fig. 3.3; change in $\Lambda_F(t)$). The two other methods for the detection of a change in the trend of the transmission intensity, although less sensitive in this application, show about the same results (fig. 3.4). For the detection of periods of change in the area of Kilinochchi, the p-value function of the R-design is used (fig. 3.3; change in $\Lambda_R(t)$). The results of the F-design and R-design analysis appear to be practically the same. If malathion was more effective than DDT then a decreasing annual trend in $\Lambda_s(t)$ and SPR has to be expected which starts already before the actual introduction of malathion. The results of the analysis of the parasitological data as well as the serological data are indicating such a downward trend, providing evidence for the effectiveness of malathion in the Kilinochchi district.

3.5.5 Spatial variation

To detect spatial variation in the trend of transmission, an overall Chi-square test described in section 3.3.3.1 has been proposed as a preliminary test. If spatial variation was not detectable with this test then the combination of the 8 subpopulations was considered homogeneous. The obtained χ^2 -value, however, was strongly significant ($\chi^2=82.56$, degrees of freedom 35, $p=10^{-5}$). The χ^2 variable is the sum of variables χ_j^2 ($j=1, \dots, 5$) where χ_j^2 is the Chi-square statistic for the comparison of the seropositivity rates of age group A_j in the 8 sub-areas. It appears that the main contributions to the χ^2 value are due to the χ_4^2 and χ_5^2 values ($\chi_1^2=2.14$, $\chi_2^2=16.34$, $\chi_3^2=12.40$,

$\chi_4^2=28.10$, $\chi_5^2=23.58$). Without doubt the seropositivity rates in the sub-areas are not equal for the age-group 3.5-4.5 yr and 4.5-5.5 yr respectively (χ_4^2 , $p=2.10^{-4}$; χ_5^2 , $p=10^{-3}$).

It might seem as if these results are inconsistent with the obtained estimates $\hat{\sigma}_\lambda(t)$ of the spatial variation in the trend of transmission (fig. 3.5, $\hat{\sigma}_\lambda(t)$ is the square root of the spatial variance (2)). Although χ_4^2 and χ_5^2 are clearly significant, the estimate $\hat{\sigma}_\lambda(t)$ of the spatial variation in period 3.5-5.0 yr before survey date is negative. These results seem to be contradictory. It would, however be erroneous to argue that a statistically significant difference in seropositivity rates for age-group A_j implies a spatial variation in the trend of transmission A_j years before survey date. It is obvious that a significant χ_j^2 value indicates a spatial variation in the trend of transmission provided that the fading of antibodies can be neglected. This spatial variation, however, may have happened at any period between survey date and A_j years before survey date. Moreover a spatial variation in the trend of transmission A_j years before survey date does not imply a difference in cumulative inoculation rates for age group A_j at survey date. Hence the χ_j^2 components of the overall Chi-square test are not suitable to explore the spatial variance further, as indicated they may even be misleading. Instead estimates have been given of the square root of a) the spatial variance in the trend-values (σ_λ^2) and b) the spatial variance in the rate of change of the trend-values (σ_Y^2) (see fig. 3.5). Further study may result in the development of a test for detecting spatial variation which is based on the estimator $\hat{\sigma}_\lambda^2(t)$.

There are indications for a spatial variation in the trend-values $\lambda_s(t)$ in period 0.8-1.9 yr, 2.9-3.3 yr before survey date; outside these periods the variation in $\hat{\lambda}_s(t)$ is completely explained by statistical fluctuations due to sampling ($\hat{\sigma}_{F,\lambda}^2$ is negative, fixed design). With respect to the spatial

variation in the rate of change in the trend of transmission, $\hat{\sigma}_Y^2(t)$ is positive in period 1.1-1.3 yr, 2.4-2.5 yr and period 3.5-3.8 yr before survey date (fig. 3.5; $\hat{\sigma}_Y(t)$ is the square root of the spatial variance (2)). The estimates of the total variance and the spatial variance in the Kilinochchi district (random design) are $n/(n-1)=8/7$ times the corresponding estimates in the eight combined sub-areas. However, the proportion explained variance in $\hat{\lambda}_s(t)$ and $\hat{\gamma}_s(t)$ respectively ($E_{\lambda}(t)$ and $E_{\gamma}(t)$, see fig. 3.5) is the same for both designs.

As stated in section 3.3.3.4 there exists a close relationship between the proportion explained variance in $\hat{\gamma}_s(t)$ due to sampling and the results of the statistical tests for the detection of a change in the trend of transmission in the eight combined sub-areas and the whole Kilinochchi district (fixed design and random design respectively). In period 0.0-1.1 yr, 1.3-2.4 yr, 2.5-3.5 yr, 3.8-5.0 yr before survey date the estimate $\hat{\sigma}_Y^2(t)$ of the spatial variance in $\gamma_s(t)$ takes on negative values. In other words the spatial variation in $\hat{\gamma}_s(t)$ is completely explained by sampling errors in these periods (fig. 3.5, $E_{\gamma}(t) > 1$). For these periods the p-value function $P_R(t)$ of the R-design takes on values which are lower than $P_F(t)$, the p-value function of the F-design. This implies that in these periods the hypothesis of no change in the average trend in transmission intensity is rejected sooner in Kilinochchi than in the eight combined sub-areas. In particular $P_R(t) < P_F(t) \leq 0.05$ in period 0.1-1.1 yr before survey date (see fig. 3.3), indicating that in this period before survey date there is more "statistical evidence" for a downward trend in transmission in Kilinochchi than in the 8 combined sub-areas. However, since it is questionable whether the 8 sub-areas can be considered effectively a random sample of sub-areas from Kilinochchi, it remains doubtful whether this conclusion is indeed valid for the total area of Kilinochchi.

The main factor responsible for biased results is probably the factor "fading of antibodies". Two related questions might be further studied:

- How insensitive, or in other words how robust, are the models of chapter 2 and 3 to departures from the assumption 'no fading of antibodies'.
- How could this bias be corrected?

The next chapter is concerned with these questions.

3.6 SUMMARY

A stochastic model is introduced for the determination of the trend of malaria transmission in a non-homogeneous population. The model takes into account the spatial-dependent and time-dependent risk of receiving inoculations and is based on estimates of the cumulative inoculation rates obtained from one cross-sectional survey. The model can be considered a generalization of the time-dependent transmission model described in the previous chapter. The non-homogeneous population is divided into m subpopulations; each subpopulation is assumed to be homogeneous. The (weighted) mean $\lambda_s(t)$ of the spline-trend functions $\lambda_s(t)$ in the subpopulations is used as a measure of the average trend in transmission intensity. The model contains parameters to measure the spatial variation in the trend of the transmission intensity.

Two designs are considered: the fixed design (F-design) and the random design (R-design). In an F-design cross-sectional surveys are performed in all subpopulations. In an R-design cross-sectional surveys are only carried out in a (weighted) random sample of subpopulations chosen from the m subpopulations into which the non-homogeneous population has been divided. For the F-design as well as the R-design a "trend-change test" is proposed for the detection of a change in the average trend of the transmission intensity. In case of an

unweighted analysis a relationship is presented between both the p-value functions of the tests and that part of the variation in the rate of change of the transmission trends $\hat{\lambda}_s(t)$, which is explained by sampling fluctuations.

Furthermore a "trend-value test" and a "trend-change test" is introduced for the comparison of the trend of transmission in two homogeneous populations.

As an illustration of the application of the model serological data from Kilinochchi, an area in the north of Sri Lanka, were used to assess the trend in transmission intensity in the years 1974-1978. As in chapter 2, it has to be assumed that evidence of exposure to infection is definite for the age band covered by the data i.e. in the present application no fading of antibodies is assumed in children less than 6 years old. The trend derived from the serological data obtained from one cross-sectional survey carried out in September-October 1978, sample size 1,678, is compared with the laboratory confirmed malaria cases for the years 1973-1978 collected in the same area (examination of 14,943 blood smears). The spline-trend in transmission intensity derived from serological data is similar to the annual trend in the quarterly slide positivity rates. This provides evidence that the model might produce valid results with respect to the detection of changes in the trend of transmission.

One year before the survey data of the cross-sectional serological survey DDT was replaced by malathion. Since the parasitological data as well as the serological data show an annual downward trend in transmission intensity after the middle of 1977, the results provide an indication for the effectiveness of malathion in the area of Kilinochchi.

CORRECTION FOR FADING OF ANTIBODIES AND SENSITIVITY AND SPECIFICITY OF THE SEROLOGICAL TEST

4

4.1 INTRODUCTION

In chapter 2 a time-dependent transmission model was introduced for the assessment of the trend of malaria intensity in a homogeneous population. In analogy with the model of Draper, Toller and Carpenter (1972) it was presupposed in that chapter that the serological survey provides the means to separate those individuals who have already experienced a parasitaemia from those who have not. It is not true, however, that circulating anti-malaria antibodies remain detectable for life, unless antigenic stimulation continues, which can be due to a chronic parasitaemia, to a parasitaemia following a true relapse or to a fresh reinfection. Hence it cannot be claimed that 'negative in a serological test' is equivalent to 'never infected'. Disregarding the factors of fading of antibodies, the sensitivity as well as the specificity of the serological test will induce a time-

dependent bias in the estimation of the trend in transmission intensity. The methodology to correct for this bias is the subject of this chapter.

Sero-epidemiological data from Mauritius, already analysed in chapter 2 without correction for fading of antibodies, are used again to illustrate the application of this method of correction in practice.

4.2 THE MATHEMATICAL MODEL

4.2.1 Introduction

In chapter 2 the spline approximations $\lambda_s(t)$ and $\gamma_s(t)$ have been introduced to describe the value and the rate of change in the trend of the transmission intensity t years before survey date. Disregarding the three factors of fading of antibodies, the sensitivity as well as the specificity of the serological test in the estimation of $\lambda_s(t)$ and $\gamma_s(t)$ implies that the estimates of these functions are erroneously based on the seropositive age related fractions. The result will be a time-dependent bias in the estimated transmission functions. In order to account for this bias first a clear distinction is made between the seropositivity rate and the cumulative inoculation rate,

$p^a(A) = \text{seropositivity rate}$
= proportion of individuals of age A who are
serologically positive¹⁾ - as measured by the test -
at the moment of blood collection.

¹⁾ For a "definition" of serologically positive see page 114

$p(A)$ = cumulative inoculation rate
= proportion of individuals of age A who have
experienced at least one infection with micro-
organism m in their life¹⁾.

The "transmission functions" based on $p^a(A)$ instead of $p(A)$ are called "antibody related transmission functions", notation $\lambda^a(t)$ and $\gamma^a(t)$ respectively; correspondingly $\lambda_s^a(t)$ and $\gamma_s^a(t)$ are used to describe their spline approximations, and $\hat{\lambda}_s^a(t)$ and $\hat{\gamma}_s^a(t)$ to indicate the estimates of these.

The epidemiological interpretation of the antibody related transmission functions $\lambda^a(t)$ and $\gamma^a(t)$ depends on the relationship of $\lambda^a(t)$ with $\lambda(t)$ and of $\gamma^a(t)$ with $\gamma(t)$. These relationships depends i.a. on the factor of fading of antibodies. The course of the fading of antibodies is complicated and confounded with the sensitivity and specificity of the serological test. Therefore the concepts of past-sensitivity and past-specificity of a serological test are introduced with account for the fading of antibodies as well as for the sensitivity and specificity.

If the past-sensitivity is 1, it means that there is no loss in antibody detectability. That is the hypothetical situation as described in chapter 2 of this study. The past-specificity can be estimated, but no quantitative information is available about the actual value of the past-sensitivity. Therefore a study was made in order to assess the effect of several assumptions, with regard to the past-sensitivity, on the interpretation of the antibody related transmission function $\lambda^a(t)$. The results of this theoretical study have been used for the development of a procedure by which a downward trend of the transmission intensity can be detected. This procedure takes into account the value and rate of change of the past-sensitivity.

¹⁾ In this study an infection with micro-organism m is equated with malaria

4.2.2 Past-sensitivity η and past-specificity ξ of a serological test

When a serological test is used to separate those individuals who have already experienced a parasitaemia from those who did not, then concepts are needed to describe the effectiveness of such a procedure. Moreover we need concepts which may be used to relate the antibody related transmission function $\lambda^a(t)$ to the transmission function $\lambda(t)$ in a mathematically convenient way. The past-sensitivity and past-specificity functions suit these purposes. In order to differentiate sensitivity from past-sensitivity and specificity from past-specificity, the formal definitions of the sensitivity and specificity of a serological test are recalled here.

The sensitivity of a serological test is normally described as the proportion of positive test results among the sera that should give a positive reaction, and the specificity as the proportion of negative test results among the sera that should not give a positive reaction. It seems logical to assume that the sera that contain specific antibodies react positively in the serological test and that sera without these antibodies do not react in that test. Accordingly the sensitivity and the specificity of a serological test are formally defined as¹⁾

$$\text{sensitivity } \eta^a = p[s^+ | a_m] \quad \text{specificity } \xi^a = p[s^- | a_m^c], \quad (4.1)$$

where s^+ = serological test result is positive

a_m = antibodies specific for micro-organism m are present at the moment of blood collection.

The symbols s^- respectively a_m^c denote the complementary situations of s^+ respectively a_m ;

¹⁾ The notation $p[|]$ refers to conditional probabilities or conditional proportions.

s^- = serological test result is negative

a_m^c = antibodies specific for micro-organism m are not present at the moment of blood collection.

The *past-sensitivity* of a serological test is defined as the proportion of individuals who are positive in the test amongst those individuals who have experienced at least one infection with micro-organism m in their life. The *past-specificity* of a serological test is accordingly defined as the proportion of individuals who are negative in the test amongst those individuals who never have experienced an infection with micro-organism m in their life. This could be written as

$$\text{past-sensitivity } \eta = p[s^+ | m] \quad \text{past-specificity } \xi = p[s^- | m^c], \quad (4.2)$$

where m = the individual has experienced at least one infection with micro-organism m in his life

m^c = the individual has not experienced an infection with micro-organism m in his life.

If specific antibodies a_m develop immediately after the infection with micro-organism m and antibody production does not drop due to a continuing antigenic stimulation then

$$m = a_m.$$

Hence in this situation past-sensitivity is equal to sensitivity, $\eta = \eta^a$, and past-specificity is equal to specificity $\xi = \xi^a$. Generally, however, antibody production can fall and therefore the factor of fading of antibodies comes in.

In order to have a clear insight into the relationship between the concepts past-sensitivity and sensitivity on the one hand and past-

specificity and specificity on the other, the degree of fading of antibodies in a population has to be defined. The degree of fading of antibodies (f) is defined as the proportion of individuals who do not have antibodies specific for micro-organism m at the moment of blood collection amongst those individuals in the population who have experienced at least one infection with micro-organism m in their life. In formula

$$\text{fading of antibodies } f = p \left[\frac{a^c}{m} \right]. \quad (4.3)$$

A Venn-diagram of the past-sensitivity, past-specificity, sensitivity, specificity of a serological test and the fading of antibodies is presented in diagram 4.1. It will be discussed in more detail on page 117.

The distribution of titres of a serological test in a population depends on the specific antibody levels in the individuals of the population as well as on the occurrence of non-specific serological reactions. The past-sensitivity η , the sensitivity η^a , the past-specificity ξ and the specificity ξ^a depend on the particular test involved and its appropriate discriminative titre level. When this level has been chosen, then the results of the serological test can be classified as negative (s^-) or positive (s^+). In order to establish a general relationship between the concepts η , η^a , ξ , ξ^a and f , which is independent of the choice of the discriminative titre level, the following assumption is made. The distribution of titres in individuals who have experienced an infection with micro-organism m but who do not have specific antibodies at the moment of blood collection (group denoted by $m - a_m$) equals the distribution of titres in those individuals who never have experienced an infection with micro-organism m in their life (group denoted by m^c). From this distribution equality of titres in group $(m - a_m)$ and group m^c follows that the proportion false positive reactions is the same for both groups.

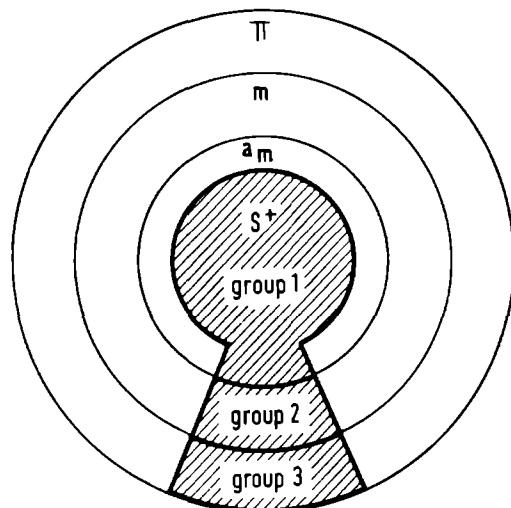
Diagram 4.1 Venn-diagram of the concepts past-sensitivity η , past-specificity ξ , sensitivity η^a , specificity ξ^a and the factor of fading of antibodies f .

m = the individual has experienced at least one infection with micro-organism m in his life

a_m = antibodies specific for micro-organism m are present at the moment of blood collection

s^+ = serological test results is positive (*shaded area*)

The symbols m^c , a_m^c and s^- denote the complements of m , a_m and s^+



Classification of serologically positive individuals
of a population Π into three groups

$$\text{past-sensitivity } \eta = p[s^+ | m]$$

$$\text{sensitivity } \eta^a = p[s^+ | a_m]$$

$$\text{past-specificity } \xi = p[s^- | m^c]$$

$$\text{specificity } \xi^a = p[s^- | a_m^c]$$

$$\text{fading of antibodies } f = p[a_m^c | m]$$

This can be written as

$$p[s^+ | m - a_m] = p[s^+ | m^c]. \quad (4.4)$$

The relationship (4.4) holds irrespective of the choice of the discriminative titre level. This implies that the serological test cannot be used to make a distinction between individuals from group $m - a_m$ and individuals from group m^c . Using (4.4) the following general relationships between the past-sensitivity η , the past-specificity ξ , the sensitivity η^a , the specificity ξ^a and the degree of fading of antibodies, the factor f have been derived¹⁾:

$$\xi = \xi^a \quad (4.5)$$

$$\eta = (1 - f)\eta^a + f(1 - \xi^a). \quad (4.6)$$

Thus under assumption (4.4) it follows that

1. The past-specificity is equal to the specificity
2. The past-sensitivity of a serological test η depends on the degree of fading of antibodies i.e. the factor f , the sensitivity η^a and the specificity ξ^a of the serological test.

When there has been no fading of antibodies, $f = 0$, then $\eta = \eta^a$. Thus the past-sensitivity and the past-specificity of a serological test can be considered generalizations of the sensitivity and specificity of a serological test.

For any serological test it can be assumed that

$$\eta^a > 1 - \xi^a.$$

¹⁾ The assumption $p[s^+ | m - a_m] = p[s^+ | m^c]$ is also a necessary condition for each of these two relationships to be true; this is illustrated in the theoretical example on page 119.

If it is supposed that $p[s^+ | m - a_m] \geq p[s^+ | m^c]$ then an analogous relationship holds between the past-sensitivity η and the past-specificity ξ i.e.

$$\eta > 1 - \xi \quad (f \neq 0). \quad (4.7)$$

It is evident that $p[s^+ | a_m] > p[s^+ | m - a_m]$. Using this inequality it is readily derived that the past-sensitivity is smaller than the sensitivity provided that the degree of fading of antibodies is not zero, i.e.,

$$\eta < \eta^a \quad (f \neq 0). \quad (4.8)$$

For the derivations of formulae and further details see Appendix 4.

The concepts defined in this section and their interrelationships are further illustrated in the remainder of this section.

Discussion of Venn-diagram 4.1

On account of theoretical considerations the individuals who are serologically positive can be classified into three groups:

group 1: individuals who have specific antimalaria antibodies, this group is denoted by $s^+ \cap a_m$.

group 2: individuals who also have had malaria in their life but fail to have specific antimalaria antibodies; the positive test result is due to a false positive reaction of the serological test. This group is denoted by $s^+ \cap (m - a_m)$.

group 3: individuals who never had malaria in their life, this group is denoted by $s^+ \cap m^c$.

The individuals in group 1 are true positives, individuals in group 3 are false positives. Individuals in group 2 have had malaria and have antibodies

that react with the crude antigen of the serological test; these reactions however belong to a category which is not specific for the malaria antigenic determinants in stricto sensu. The individuals in group 2 are therefore false positives from a serological point of view. In this study, however, a serological test is used for the separation of individuals who have experienced a parasitaemia in their life from those who have not. Therefore in this context the seropositive individuals in group 2 can also be regarded as true positives.

The theoretical classification of serologically positive individuals in three groups and the relations (4.5) to (4.8) are illustrated in the following "numerical" example.

A population Π is considered which consists of 1000 individuals; 450 individuals are serologically positive on the day of the survey. It is assumed that 600 individuals have had malaria in their life and that 400 of these individuals still have antibodies at the moment of blood collection. The 450 seropositive individuals are theoretically classified into three groups (see diagram 4.1). It is supposed that:

- 360 of these individuals actually have antibodies (group 1)
- 30 of these individuals have had malaria in their life, however, they do not have antibodies specific for malaria at the moment of blood collection (group 2)
- 60 of these individuals never had malaria (group 3)

Group 2 and group 3 represent the false positive reactions.

Using these hypothetical figures the sensitivity, the specificity, the past-sensitivity, the past-specificity and the degree of fading of antibodies are easily calculated. Let $N(\Pi)$ be the total size of the population ($N(\Pi) = 1000$) and let $N(x)$ be the number of individuals in the population having

characteristic x. Using the notation of this section the figures of the example can be rendered by

$$N(m) = 600, N(a_m) = 400, N(s^+) = 450,$$

$$N(s^+ \cap a_m) = 360, N(s^+ \cap (m - a_m)) = 30, N(s^+ \cap m^c) = 60.$$

From the definitions of η^a , ξ^a , η , ξ and f (see also diagram 4.1) immediately follows:

$$\text{sensitivity } \eta^a = p[s^+ | a_m] = 360/400 = 90\%$$

$$\text{specificity } \xi^a = p[s^- | a_m^c] = (600-90)/600 = 85\%$$

$$\text{past-sensitivity } \eta = p[s^+ | m] = (360+30)/600 = 65\%$$

$$\text{past-specificity } \xi = p[s^- | m^c] = (400-60)/400 = 85\%$$

$$\text{fading of antibody } f = p[a_m^c | m] = (600-400)/600 = 33\frac{1}{3}\%$$

It is easily seen that $\xi = \xi^a$ and $\eta = (1 - f)\eta^a + f(1 - \xi^a)$. These relationships, however, are only true if it is assumed that relation (4.4) holds i.e. the proportions of false positive reactions in group $(m - a_m)$ and group m^c are the same (in this example 15%). This is a necessary condition for the relationships (4.5) and (4.6) to be generally true. If for instance in the above example the number of false positives in group 2 and group 3 would be 50 and 40 respectively instead of 30 and 60 then it would follow that

$$p[s^+ | m - a_m] = 50/(600-400) = 25\%, p[s^+ | m^c] = 40/400 = 10\%.$$

The sensitivity η^a , the specificity ξ^a of the serological test as well as the degree of fading of antibodies f remain unchanged. The past-sensitivity η and the past-specificity ξ , however, take on larger values: $\eta = (50 + 360)/600 = 68\frac{1}{3}\%$ and $\xi = (400-40)/400 = 90\%$.

4.2.3 Instantaneous correction for the factor past-sensitivity

4.2.3.1 Introduction

In order to avoid ambiguity in the mathematical description and epidemiological interpretation of the course of the "transmission functions" $\lambda^a(t)$ and $\lambda(t)$, emphasis is placed on the conventions regarding the description of time and age (see also Appendix 1, section 1.4).

The symbol t in the functions $\lambda^a(t)$, $\lambda(t)$ and their derivatives $\gamma^a(t)$ and $\gamma(t)$ denotes a time point t years before survey date ($t > 0$). The epidemiological interpretation of these functions, however, is in the opposite direction i.e. from the past to the survey date. Accordingly a negative sign of $\gamma^a(t)$ is interpreted as an increase in $\lambda^a(t)$ and a positive sign of $\gamma^a(t)$ corresponds with a decrease in $\lambda^a(t)$. The same convention holds for the interpretation of the sign of $\gamma(t)$. Furthermore the symbol A may refer to a time point A years before survey date or to age A (years), for instance:

$\lambda^a(A) =$ value of the antibody related transmission function $\lambda^a(t)$ in a time point A years before survey date,

$\eta(A) =$ past-sensitivity of the serological test for individuals of age A (years).

The course of the age related functions are described and interpreted in the conventional way, e.g. $\frac{dn}{dA} > 0$ means the past-sensitivity $\eta(A)$ increases with age.

The interpretation of the antibody related transmission function $\lambda^a(t)$ in this section is based on some additional assumptions which extend the series stated in chapter 2. These assumptions are:

- The seropositivity rate $p^a(A)$ increases with age A
- The past-specificity ξ of the serological test is 1

In section 4.2.3.3 and 4.2.3.4. certain additional assumptions are made with respect to the past-sensitivity η of the serological test. In case ξ is less than 1, a correction method for the simultaneous effect of η and ξ on the interpretation of $\lambda^a(t)$ is presented in section 4.4.

4.2.4.3 The sign disturbance coefficient

The value of the transmission function $\lambda(t)$ and the antibody related transmission function $\lambda^a(t)$ in a time point A years before survey date is equal to

$$\lambda(A) = \frac{\frac{dp(A)}{dA}}{1-p(A)} \quad \lambda^a(A) = \frac{\frac{dp^a(A)}{dA}}{1-p^a(A)} . \quad (4.9)$$

The assumption of an increasing seropositivity rate $p^a(A)$ with age, ensures that the antibody related transmission function $\lambda^a(t)$ takes on positive values. The interpretation of the course of $\lambda^a(t)$ depends on the relationship between the seropositivity rate $p^a(A)$ and the cumulative inoculation rate $p(A)$. This relationship is easily established employing the concepts of age specific past-sensitivity $\eta(A)$ and age-specific past-specificity $\xi(A)$ of the serological test.

As $\eta(A) = p_A[s^+|m] =$ past-sensitivity η for age A,
 $\xi(A) = p_A[s^-|m^c] =$ past-specificity ξ for age A,
 $p^a(A) = p_A[s^+] =$ seropositivity rate for age A,
 $p(A) = p_A[m] =$ cumulative inoculation rate for age A,

and $p_A[s^+] = p_A[s^+|m] p_A[m] + p_A[s^+|m^c] p_A[m^c]$ it is seen that
 $p^a(A) = \eta(A) p(A) + [1-\xi(A)] [1-p(A)]. \quad (4.10)$

If the past-sensitivity and the past-specificity of the serological test would be both equal to 1, $\eta(A) = 1$ and $\xi(A) = 1$, then $p^a(A) = p(A)$ and accord-

ingly the antibody related transmission function $\lambda^a(t)$ would be equal to the real transmission function $\lambda(t)$. It is complicated to study the simultaneous effect of the two age-dependent variables $\eta(A)$ and $\xi(A)$ on the estimation and testing procedures introduced in chapter 2 and to derive the relationship between $\lambda^a(t)$ and $\lambda(t)$. If, however, a high discriminative titre level of the serological test is selected, then it may be assumed that the past-specificity is 1 ($\xi(A)=1$). The relationship between $p^a(A)$ and $p(A)$ is then (see 4.10)

$$p^a(A) = \eta(A) p(A). \quad (4.11)$$

The past-sensitivity $\eta(A)$ is determined in this situation by the degree of fading of antibodies $f(A)$ and the sensitivity $\eta^a(A)$ of the serological test. From (4.6) follows

$$\eta(A) = [1 - f(A)] \eta^a(A). \quad (4.12)$$

The general relationship between the value of the transmission function $\lambda(t)$ and the value of the antibody related transmission function $\lambda^a(t)$ in a time point A years before survey date is (see Appendix 4)

$$\lambda(A) = \left[\frac{1 - \eta(A)}{\eta(A) - p^a(A)} + 1 \right] \left[\lambda^a(A) - \frac{1}{\eta(A)} \frac{d\eta}{dA} \frac{p^a(A)}{1 - p^a(A)} \right]. \quad (4.13)$$

From (4.13) follows that the interpretation of the value of the antibody related transmission function $\lambda^a(t)$ in a time point A years before survey date depends on:

- the seropositivity rate $p^a(A)$,
- the value of the past-sensitivity $\eta(A)$ and
- the rate of change of the past-sensitivity $\frac{d\eta}{dA}$.

These factors do not vary independently. From (4.11) follows immediately that the seropositivity rate is a lower bound for the past-sensitivity. Furthermore

as $\lambda(A)$ is always positive, it is easily derived from (4.9) and (4.13) that the relative rate of change of the seropositivity rate is an upperbound for the relative rate of change of the past-sensitivity. In formulae

$$\eta(A) > p^a(A), \quad (4.14)$$

$$\frac{1}{\eta(A)} \frac{d\eta}{dA} < \frac{1}{p^a(A)} \frac{dp^a}{dA}. \quad (4.15)$$

The relationship between the rate of change of $\lambda(t)$ and the rate of change of $\lambda^a(t)$ in a time point A years before survey date is given by

$$\gamma(A) = \left[\frac{1 - \eta(A)}{\eta(A) - p^a(A)} + 1 \right] \left[\gamma^a(A) + \Delta(A) \right], \quad (4.16)$$

where $\Delta(A)$ is a special function which is defined in Appendix 4.

As $\eta(A) > p^a(A)$, see (4.14), it follows that the sign of $\gamma(A)$ equals the sign of $\gamma^a(A) + \Delta(A)$. Therefore the term $\Delta(A)$ is called "*the sign disturbance coefficient*". The formula for $\Delta(A)$ is rather complicated. Here it is only stated that the sign disturbance coefficient $\Delta(A)$ depends on the following related five variables (A fixed):

- the seropositivity rate $p^a(A)$
- the value of the antibody related transmission function $\lambda^a(A)$
- the value of the past-sensitivity $\eta(A)$
- the rate of change of the past-sensitivity $\frac{d\eta}{dA}$
- the curvature of the past-sensitivity $\frac{d^2\eta}{dA^2}$.

In mathematical notation this can be rendered as follows

$$\Delta(A) = \Delta(p^a(A), \lambda^a(A), \eta(A), \frac{d\eta}{dA}, \frac{d^2\eta}{dA^2}). \quad (4.17)$$

The variables $p^a(A)$ and $\lambda^a(A)$ can be estimated from the data. The value $\eta(A)$,

its rate of change $\frac{dn}{dA}$ and its curvature $\frac{d^2n}{dA^2}$, however, are virtually unknown. Therefore it is impossible to assess the value of the sign disturbance coefficient $\Delta(A)$ taking into consideration all five variables. One way out of this problem is to disregard one or more of these variables. In section 4.2.3.3 the variables $p^a(A)$, $\lambda^a(A)$, $n(A)$ are taken into account and the variables $\frac{dn}{dA}$ and $\frac{d^2n}{dA^2}$ are disregarded. In section 4.2.3.4 the rate of change $\frac{dn}{dA}$ of the past-sensitivity is additionally taken into account while the curvature $\frac{d^2n}{dA^2}$ is still ignored.

The sign of $\gamma^a(A) + \Delta(A)$ is studied by varying the past-sensitivity $n(A)$ and its rate of change $\frac{dn}{dA}$. The proposed interpretation of the course of the antibody related transmission function $\lambda^a(A)$ in a time point A years before survey date in the next two sections (4.2.3.3 and 4.2.3.4) is based on the following :

1. If $\gamma^a(A) + \Delta(A)$ can attain the value zero then it is decided that the course of the real transmission function $\lambda(t)$ is uncertain in a time point A years before survey date.
2. If $\gamma^a(A) + \Delta(A)$ can not attain the value zero then it is decided that the course of the antibody related transmission function $\lambda^a(t)$ reflects a real change in $\lambda(t)$, A years before survey date.

4.2.3.3 Past-sensitivity is independent of age, is constant and its level is unknown

The general relationship between the transmission function $\lambda(t)$ and the antibody related transmission function $\lambda^a(t)$ in a time point A years before survey date is expressed by formula (4.13). If the past-sensitivity is independent of age ($\frac{dn}{dA} = 0$) then it follows that

$$\lambda(A) = \left[\frac{1 - n}{n - p^a(A)} + 1 \right] \lambda^a(A). \quad (4.18)$$

From (4.18) it follows that the relative bias $(\lambda^a(A) - \lambda(A))/\lambda(A)$ in the antibody related transmission function A years before survey date is

$$= \frac{1 - \eta}{1 - p^a(A)}. \quad (4.19)$$

If the past-sensitivity does not change with age, then the seropositivity rate $p^a(A)$ increases with age (see (4.11)). Therefore it is readily seen from (4.19) that a constant, unknown past-sensitivity ($\eta \neq 1$) will cause a relative bias in the estimation of the transmission function which increases from the survey date to the past. In other words: for time points A belonging to an earlier period before survey date, the value of the antibody related transmission function $\hat{\lambda}^a(A)$ becomes increasingly less reliable as an estimate of the value of the real transmission intensity $\lambda(A)$.

The relationship between the rate of change of the transmission function ($\gamma(A)$) and the rate of change of the antibody related transmission function ($\gamma^a(A)$) in a time point A years before survey date is

$$\gamma(A) = \left[\frac{1 - \eta}{\eta - p^a(A)} + 1 \right] \left[\gamma^a(A) + \Delta(A) \right]. \quad (4.20)$$

From the general formula for the sign disturbance coefficient presented in Appendix 4 (formula (4.23)) and $\frac{d\gamma}{dA} = \frac{d^2\eta}{dA^2} = 0$ it follows immediately that

$$\Delta(A) = \frac{1 - \eta}{\eta - p^a(A)} \lambda^a(A)^2. \quad (4.21)$$

Since $\eta > p^a(A)$, see (4.14), it follows that $\Delta(A)$ is strictly positive ($\eta \neq 1$: $\Delta(A) > 0$). Therefore a decreasing antibody related transmission function ($\gamma^a(A) > 0$) will always reflect a real decrease ($\gamma(A) > 0$), irrespective of the constant value of the unknown past-sensitivity η . The interpretation of an increasing antibody related transmission function, however, depends clearly on the actual value of the past-sensitivity. If the past-sensitivity η would

be larger than an age related function $\eta_c(A)$, $\eta > \eta_c(A)$, then the real transmission function $\lambda(t)$ is increasing too. However if the past-sensitivity η would be smaller than $\eta_c(A)$, then the transmission function $\lambda(t)$ is actually decreasing instead of increasing as suggested by the course of $\lambda^a(t)$. The critical function $\eta_c(A)$ is a mathematically derived function, it has been called the critical past-sensitivity and is defined by

$$\eta_c(A) = \frac{\lambda^a(A)^2 - \gamma^a(A) p^a(A)}{\lambda^a(A)^2 - \gamma^a(A)} \quad (\gamma^a(A) < 0). \quad (4.22)$$

The main conclusions of this section are summarized in diagram 4.2. For further details see Appendix 4.

4.2.3.4 Past-sensitivity changes with age; linear correction

In the previous section the effect of a constant past-sensitivity on the interpretation of the value and the course of $\lambda^a(t)$ has been studied. The past-sensitivity, however, is not constant, generally its level will change with age. Therefore the interpretation of $\lambda^a(t)$ in a time point A years before survey date does not only depend on $\eta(A)$ but also on $\frac{d\eta}{dA}$. From the general relationship between $\lambda(A)$ and $\lambda^a(A)$, see (4.13), it is readily verified that

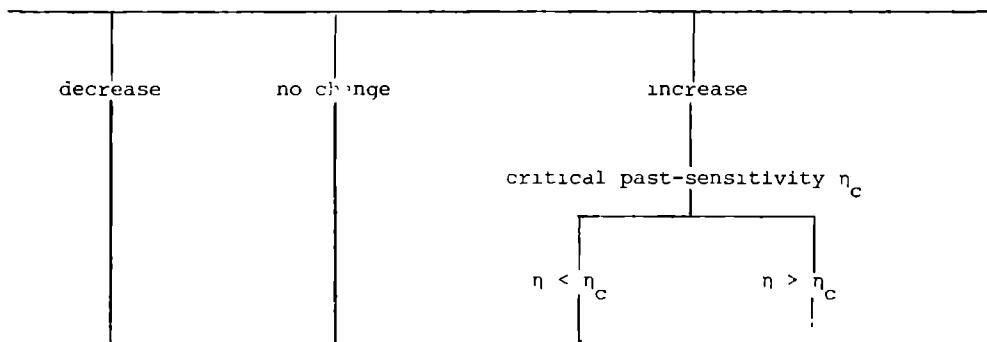
$$\lambda^a(A) < \lambda(A) \iff \frac{\frac{d\eta}{dA}}{\eta(A)(1 - \eta(A))} < \frac{\frac{dp^a}{dA}}{p^a(A)(1 - p^a(A))}. \quad (4.23)$$

It was presupposed, that the seropositivity rate $p^a(A)$ is increasing with age ($\frac{dp^a}{dA} > 0$). Consequently a decreasing past-sensitivity, a constant past-sensitivity and even a "small" increasing past-sensitivity would still cause in such a situation an underestimation of the real level of the transmission intensity $\lambda(A)$ ($\lambda^a(A) < \lambda(A)$). If, however, the rate of increase of the past-sensitivity $\eta(A)$ with age is sufficiently large, then $\lambda^a(A)$ is larger than

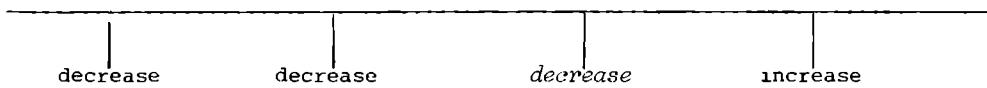
Diagram 4.2 Interpretation of the course of the antibody related transmission function $\lambda^A(t)$ at a time point A years before survey date.

Past-sensitivity η is independent of age, is constant and its value is unknown; past-specificity $\xi(A) = 1$.

course of the antibody related transmission function $\lambda^A(t)$



course of the transmission function $\lambda(t)$



legenda: The course of $\lambda^A(t)$ and $\lambda(t)$ is described from the past to the survey date. The symbol A is suppressed for clarity of notation. The critical past-sensitivity η_c is defined by (4.22).

the value of the real transmission function $\lambda^a(A)$.

In order to interpret the course of the antibody related transmission function, A years before survey date, the general relationship between $\gamma^a(A)$ and $\gamma(A)$ is used (see (4.16) and (4.17)). In the following it is assumed that the curvature of the past-sensitivity can be disregarded ($\frac{d^2\eta}{dA^2} = 0$), only the value $\eta(A)$ and its rate of change $\frac{d\eta}{dA}$ are taken into account. Three situations have been studied:

- a) Past-sensitivity $\eta(A)$ decreases with age, its value and the rate of decrease are unknown;
- b) Past-sensitivity $\eta(A)$ increases with age, again its value and the rate of increase are unknown;
- c) Past-sensitivity $\eta(A)$ changes with age, its value, the rate of change and the direction of change are unknown.

These three situations cover all possibilities in terms of linear correction. From these three types of correction number 3 is specified in the weakest way and therefore has potentially the greatest effect on the interpretation of the course of the antibody related transmission function $\lambda^a(t)$. The interval of all values that the sign disturbance coefficient $\Lambda(A)$ may attain, has been determined for each of these 3 situations. The interpretation of the course of $\lambda^a(t)$, A years before survey date, is based on the procedure described at the end of section 4.2.3.2.

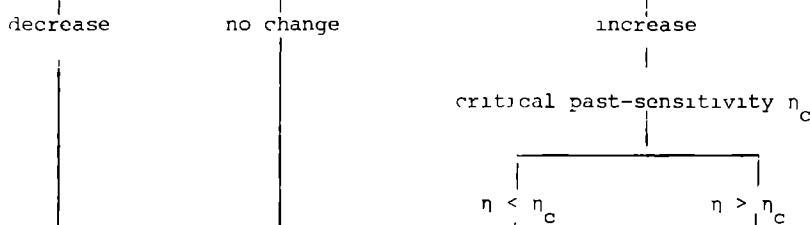
- a) Past-sensitivity $\eta(A)$ decreases with age, its level and the rate of decrease are unknown.

The main conclusions are summarized in diagram 4.3^a. As in the situation of a constant past-sensitivity (see diagram 4.2), a decrease in $\lambda^a(t)$ always indicates a real decrease. Furthermore an increase in $\lambda^a(t)$ still means a

Diagram 4.3^a Interpretation of the course of the antibody related transmission function $\lambda^A(t)$ at a time point A years before survey date.

Past-sensitivity $\eta(A)$ decreases with age, its value and the rate of the decrease are unknown; past-specificity $\xi(A) = 1$.

course of the antibody related transmission function $\lambda^A(t)$



course of the transmission function $\lambda(t)$



legenda: The course of $\lambda^A(t)$ and $\lambda(t)$ is described from the past to the survey date. The symbol A is suppressed for clarity of notation. The critical past-sensitivity n_c is defined by (4.22).

decrease in $\lambda(t)$ if $\eta(A) < \eta_c(A)$ with $\eta_c(A)$ defined by (4.22). In contrast, however, with diagram 4.2, an increase in the transmission function $\lambda(t)$ cannot be detected anymore.

- b) Past sensitivity $\eta(A)$ increases with age, its level and the rate of the increase are unknown.

The theoretical conclusions are summarized in diagram 4.3^b. A distinction is made between the interpretation of a decreasing and increasing antibody related transmission function $\lambda^a(t)$.

- Antibody related transmission function $\lambda^a(t)$ is decreasing ($\gamma^a(A) > 0$) -

Diagram 4.3^b shows that a "sufficiently" large decrease still indicates a real decrease. That is, if the rate of decrease $\gamma^a(A)$ is larger than a mathematical derived function $u(A)$, then it can be concluded that $\lambda(t)$ is decreasing A years before survey date. On the other hand if the rate of the decrease $\gamma^a(A)$ is too small, smaller than a mathematically derived function $\ell(A)$, then the course of the real transmission function is uncertain. In case the rate of decrease $\gamma^a(A)$ takes on a value between $\ell(A)$ and $u(A)$ the interpretation of the decrease depends on the actual value of the past-sensitivity for age A as compared with the value of an age related function $\eta_c(A)$ defined below.

The formulae for the lower bound $\ell(A)$, the upper bound $u(A)$ and the age related function $\eta_c(A)$, called critical past-sensitivity, are respectively:

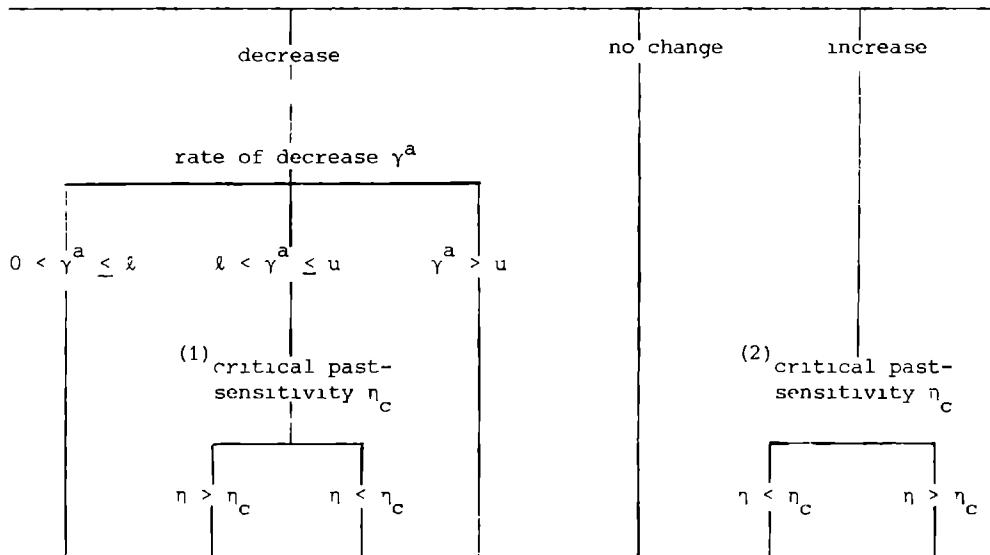
$$\ell(A) = \lambda^a(A)^2 \quad (4.24)$$

$$u(A) = \frac{\lambda^a(A)^2}{p^a(A)(2 - p^a(A))} \quad (4.25)$$

Diagram 4.3^b Interpretation of the course of the antibody related transmission function $\lambda^a(t)$ at a time point A years before survey date.

Past sensitivity η^A) increases with age, its value and the rate of the increase are unknown; past-specificity $\xi(t) = 1$.

course of the antibody related transmission function $\lambda^a(t)$



course of the transmission function $\lambda(t)$

uncertain uncertain decrease decrease uncertain uncertain increase

legenda: The course of $\lambda^a(t)$ and $\lambda(t)$ is described from the past to the survey date. The symbol A is suppressed for clarity of notation. The symbols l and u (lower bound and upper bound) are defined by (4.24) and (4.25).

(1) n_c is defined by (4.26); (2) n_c is defined by (4.22).

$$\eta_c(A) = \frac{\lambda^a(A)^2 - \gamma^a(A)p^a(A)}{(1 + p^a(A)^{-1}) \lambda^a(A)^2 - 2\gamma^a(A)} \text{ for } \lambda(A) < \gamma^a(A) \leq u(A). \quad (4.26)$$

The formula $\eta_c(A)$ presented here and defined for $\lambda(A) < \gamma^a(A) \leq u(A)$ is different from the formula for $\eta_c(A)$ defined if $\gamma^a(A) < 0$, see (4.22). Both functions are called critical past-sensitivity because they play a comparable role in the interpretation of the course of the antibody related transmission function. The $\eta_c(A)$ with $\gamma^a(A) < 0$ is associated with the interpretation of an increase in $\lambda^a(t)$ whereas $\eta_c(A)$ defined in this section is used for the interpretation of a decrease in $\lambda^a(t)$. For further details with respect to the mathematical derivation of the critical past-sensitivity $\eta_c(A)$ see Appendix 4.

- Antibody related transmission function $\lambda^a(t)$ is increasing ($\gamma^a(A) < 0$) -

The interpretation of an increasing antibody related transmission function $\lambda^a(t)$, A years before survey date, depends on the result of the comparison of the past-sensitivity $\eta(A)$ for age A with the critical past-sensitivity $\eta_c(A)$ defined by (4.22). Hence an increase in $\lambda^a(t)$ only reflects a real increase if the past-sensitivity $\eta(A)$ is larger than the critical past-sensitivity $\eta_c(A)$.

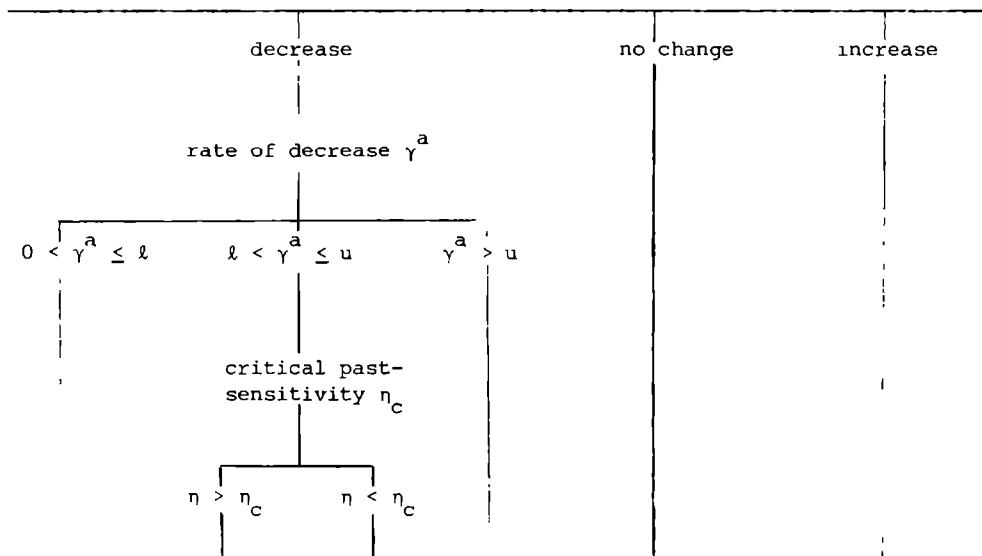
c. Past-sensitivity $\eta(A)$ changes with age, its level, the rate of change and the direction of change are unknown.

The interpretation of the course of the antibody related transmission function in this situation is summarized in diagram 4.4. This diagram combines the results of diagram 4.3^a and 4.3^b i.e. it is concluded that the real transmission $\lambda(t)$ decreases (increases) if it decreases (increases) in both diagrams.

Diagram 4.4 Interpretation of the course of the antibody related transmission function $\lambda^a(t)$ at a time point A years before survey date.

Past-sensitivity $n(\lambda)$ changes with age, its value, the rate of change and the direction of change are unknown; past-specificity $\xi(\lambda) = 1$.

course of the antibody related transmission function $\lambda^a(t)$



course of the transmission function $\lambda(t)$



legenda: The course of $\lambda^a(t)$ and $\lambda(t)$ is described from the past to the survey date. The symbol A is suppressed for clarity of notation. The symbols l and u (lower bound and upper bound) are defined by (4.24) and (4.25). The critical past-sensitivity n_c is defined by (4.26).

The main conclusions are:

- An increasing antibody related transmission function $\lambda^a(t)$ does not allow any interpretation with regard to the course of the actual transmission function $\lambda(t)$. That is an increase in $\lambda^a(t)$ can always be explained by a linear course of the past-sensitivity $\eta(A)$ by age.
- More can be said if the antibody related transmission function $\lambda^a(t)$ shows a decrease. If the rate of decrease A years before survey date $\gamma^a(A)$ is sufficiently large, larger than a mathematically derived function $u(A)$ which can be estimated from the data, then this decrease reflects a real decrease. If the rate of decrease $\gamma^a(A)$ assumes a value lower than the upperbound $u(A)$ but larger than a lowerbound $l(A)$ then the critical past-sensitivity $\eta_c(A)$ defined by formula (4.26), might be used to interprete the decrease of $\lambda^a(t)$, A years before survey date.

4.2.3.8 Interpretation of the course of the antibody related transmission function

It is convenient to have one surveyable diagram which contains the essential information of the diagrams 4.2, 4.3^{a,b} and diagram 4.4 and which can be used to interprete a decreasing as well as an increasing antibody related transmission function in relation with the course of the past-sensitivity $\eta(A)$ of the serological test. Diagram 4.5 presents such a summary of these four diagrams. It clearly shows that an increase in the real transmission function $\lambda(t)$ is indicated in the following situation:¹⁾

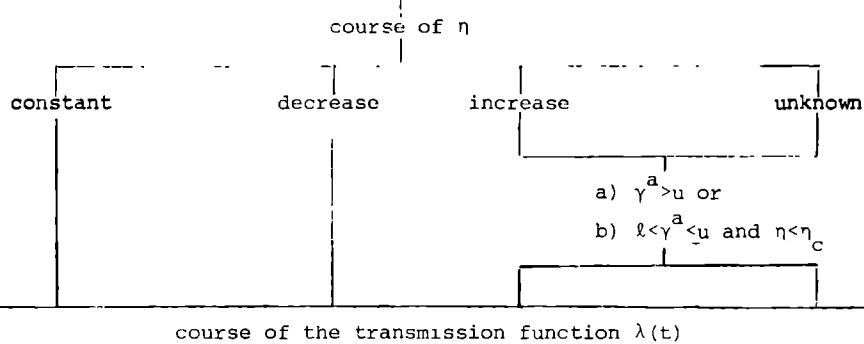
- $\lambda^a(t)$ is increasing and
- the past-sensitivity $\eta(A)$ is not decreasing with age and

¹⁾ Note: the curvature of the past-sensitivity is disregarded throughout

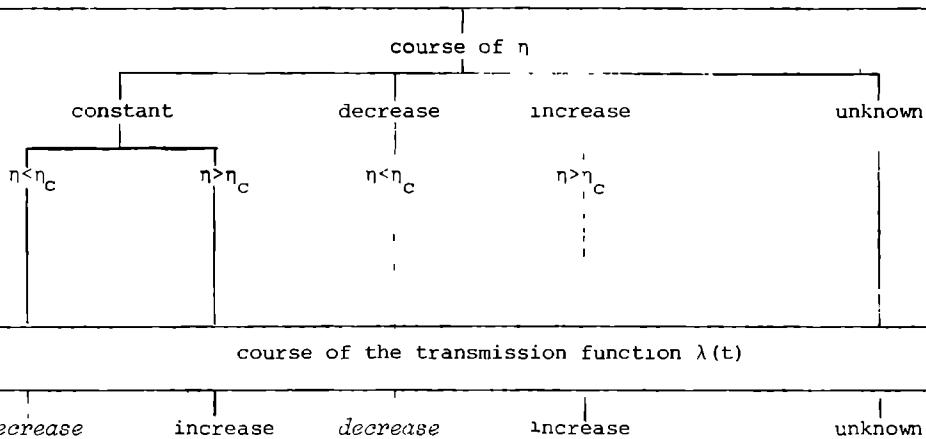
$$\left(\frac{d^2\eta}{dA^2} = 0\right).$$

Diagram 4.5 Interpretation of the course of the antibody related transmission function $\lambda^a(t)$ at a time point A years before survey date; a summary of situations based on the diagrams 4.2, 4.3^{a,b} and 4.4.

Antibody related transmission function $\lambda^a(t)$ is decreasing¹⁾



Antibody related transmission function $\lambda^a(t)$ is increasing²⁾



legenda: The course of $\lambda^a(t)$ and $\lambda(t)$ is described from the past to the survey date. The symbol A is suppressed for clarity of notation.

- 1) The critical past-sensitivity n_c is defined by (4.26).
- 2) The critical past-sensitivity n_c is defined by (4.22).

- The past-sensitivity $\eta(A)$ is larger than the critical past-sensitivity $\eta_c(A)$ defined by formula (4.22).

Therefore as the value of the past-sensitivity $\eta(A)$, its rate of change and the direction of change are unknown, an increase in $\lambda^a(t)$ at a time point A years before survey date cannot be interpreted in an objective way.

A decrease in the real transmission function $\lambda(t)$ is indicated in many situations. For instance a decreasing antibody related transmission function $\lambda^a(t)$ always implies a real decrease if the past-sensitivity $\eta(A)$ is independent of age or decreasing with age. In these two situations even an increase in $\lambda^a(t)$ may actually indicate a decrease in transmission intensity if the past-sensitivity $\eta(A)$ is smaller than the critical past-sensitivity $\eta_c(A)$ defined by formula (4.22). If $\eta(A)$ increases for age A, then a decrease in $\lambda^a(t)$ in a time point A years before survey date indicates a real decrease if one of the following two conditions is fulfilled¹⁾:

- the rate of decrease $\gamma^a(A)$ is sufficiently large ($\gamma^a(A) > u(A)$)
- the rate of decrease $\gamma^a(A)$ takes on a value between the lowerbound $\ell(A)$ and the upperbound $u(A)$ and the past-sensitivity $\eta(A)$ is smaller than the critical past-sensitivity.

Since the value of the past-sensitivity $\eta(A)$ its rate of change and the direction of change are unknown an "objective" method for the detection of a downward trend in transmission should be based on the first condition i.e.

$$\text{If } \gamma^a(A) > u(A) \text{ then } \gamma(A) > 0. \quad (4.27)$$

In the next section this condition is used for the development of a correction method for the factor of past-sensitivity $\eta(A)$.

¹⁾ The lowerbound $\ell(A)$ and the upperbound $u(A)$ are defined by the formulae (4.24) and (4.25)

4.2.4 Detection of a downward trend of the transmission intensity in a homogeneous population

The proposed procedure for the detection of a downward trend in transmission intensity consists of two steps:

step 1 Periods before survey date are determined in which the trend ($\lambda_s^a(t)$) of the antibody related transmission function is decreasing. Here the "trend-change test" - introduced in chapter 2 - is applied. Only those periods before survey date in which the spline approximation

$\hat{\lambda}_s^a(t)$ is significantly decreasing (one-sided test, $p \leq 0.05$) are analysed further in step 2 of the procedure. If the one-sided trend-change test is not significant ($p > 0.05$), then step 2 of the procedure is not performed and the decision is made that the data do not provide evidence for a downward trend in transmission intensity.

step 2 In this step a correction for the factor of past-sensitivity $\eta(A)$ is carried out. Periods found in step 1 are considered for which the spline-trend $\hat{\lambda}_s^a(t)$ is positive. A downward trend in transmission intensity is indicated in those (sub-) periods for which the following condition holds

$$\hat{\gamma}_s^a(A) > \hat{u}(A). \quad (4.28)$$

The $\hat{\gamma}_s^a(A)$ is the spline estimate of $\gamma^a(A)$ and its formula was presented in chapter 2 (2.17). The $\hat{u}(A)$ is an estimate of the upperbound $u(A)$ and is obtained by substituting appropriate estimates of $\lambda^a(A)$ and $p^a(A)$ in the formula of $u(A)$, see (4.25),

$$\hat{u}(A) = \frac{\hat{\lambda}_s^a(A)^2}{\hat{p}^a(A)(2 - \hat{p}^a(A))}. \quad (4.29)$$

In the applications the seropositivity rate $p^a(A)$ is simply estimated by linear interpolation using the seropositivity rates $\hat{p}^a(A_j)$, $\hat{p}^a(A_{j+1})$ of the two nearest age-groups ($A_j < A < A_{j+1}$).

Step 1 and step 2 of the procedure are summarized as follows. Periods before survey date in which $\hat{\lambda}_s^a(t)$ is "significantly" decreasing and in which the rate of the decrease $\hat{\gamma}_s^a(t)$ is "sufficiently" large are considered to indicate a downward trend in transmission. The method of correction in step 2 takes into account the most unfavourable situation in terms of linear correction for the factor of past-sensitivity $\eta(A)$. The proposed method of correction is therefore called *maximal linear correction (MLC)* for the factor of past-sensitivity.

If the rate of decrease $\hat{\gamma}_s^a(A)$ assumes a value between (estimates¹⁾ of) the upperbound $u(A)$ and the lowerbound $\ell(A)$, then a subjective decision with regard to the trend of the transmission intensity might be possible. If one is willing to believe that the actual unknown value of the past-sensitivity $\eta(A)$ is smaller than the estimate of the critical past-sensitivity $\eta_c(A)$, then - according to diagram 4.5 - the data still provide evidence for a downward trend of the transmission intensity. The critical past-sensitivity $\eta_c(A)$ has been estimated by (see also (4.26))

$$\hat{\eta}_c(A) = \frac{\hat{\lambda}_s^a(A)^2 - \hat{\gamma}_s^a(A) \hat{p}^a(A)}{(1 + \hat{p}^a(A)^{-1}) \hat{\lambda}_s^a(A)^2 - 2\hat{\gamma}_s^a(A)}, \quad (4.30)$$

where $\hat{p}^a(A)$ is defined as in (4.29).

This method based on the subjective comparison of $\eta(A)$ and $\hat{\eta}_c(A)$ is occasionally used in the applications.

¹⁾ Estimates of $\ell(A)$, $u(A)$, are obtained by substituting appropriate estimates of $\lambda^a(A)$ and $p^a(A)$ in (4.24) and (4.25) (see (4.29)).

In the next section the Mauritius data are used again to illustrate the application of the two-step procedure for the detection of a downward trend in transmission intensity.

4.3 ILLUSTRATION; SERO-EPIDEMOIOLOGICAL DATA FROM MAURITIUS

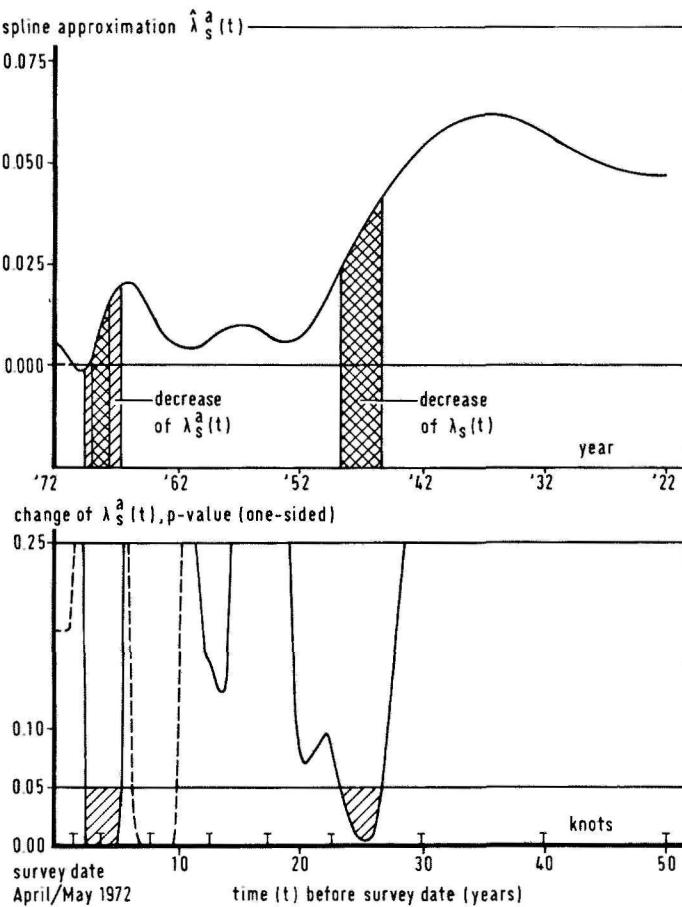
In contrast to the analysis in chapter 2 which was based on a two-sided test for detecting a change in the spline approximation $\hat{\lambda}_s^a(t)$, now the associated one-sided test is carried out (step 1 of the procedure).

Additionally the maximal linear correction (MLC) method is used in order to correct for the factor of past-sensitivity of the serological test (step 2 of the procedure). Fig 4.1 (see page 140) presents the results of the two-step procedure. This type of presentation will be used as a standard presentation in the later following applications (see chapter 5). In interpreting fig. 4.1 four parts of the figure have to be considered consecutively:

1. The graphical representation of the spline approximation $\hat{\lambda}_s^a(t)$ of the antibody related transmission function in the upper-half of the figure.
2. The one-sided p-value function in the lower-half of the figure. If the solid curve¹⁾ falls below 0.05, then the spline approximation $\hat{\lambda}_s^a(t)$ is considered to decrease in that particular point of time before survey date (single shaded area in the lower-half of the figure).
3. Those periods in which the spline approximation $\hat{\lambda}_s^a(t)$ is assumed to decrease are indicated in the upper-half of the figure by the single shaded

¹⁾ The main objective is the detection of a downward trend in transmission intensity ($\lambda_s^a(t)$). However, occasionally it is also the intention to discuss the detection of periods in which the trend of the transmission intensity might have increased; therefore the associated one-sided p-value curve (interrupted line) in the lower-half of the figure is used.

Figure 4.1 Application of the two-step procedure for the detection of a downward trend of the transmission intensity in Mauritius.



See general legenda page 247:

$\hat{\lambda}_s^a(t)$ = spline-trend of the antibody related transmission function

$\lambda_s(t)$ = spline-trend of the transmission intensity

area ///. It is noted that double shading always implies single shading.

4. Finally the correction for the factor of past-sensitivity is considered.

Subperiods of the periods indicated in part 3 are shown in the upper-half of the figure with a decrease in $\hat{\lambda}_s^a(t)$ sufficiently large to assume a decrease in the spline approximation $\lambda_s^a(t)$. These subperiods are indicated by an additional shading in the other direction \\\ .

In order to avoid an erroneous interpretation of the value of the spline approximation $\hat{\lambda}_s^a(t)$, the description of the unit of the vertical axis is deliberately not presented in the upper-half of the figure.

The analysis of the Mauritius data along these lines indicates a decrease of the spline approximation $\lambda_s^a(t)$ in two periods (step 1 of the procedure). Correction for the factor of past-sensitivity, step 2 of the procedure using the MLC method, yields two (sub-)periods, 2.8-4.8 yr and 23.6-26.9 yr before survey date, indicating a real decrease in the trend of the transmission intensity in these periods.

The following results of the analysis are less informative from a practical point of view. However, they illustrate the potential meaning of the critical past-sensitivity $n_c(A)$ for the detection of a downward or an upward trend in the transmission intensity. Table 4.1 (see page 142) presents the periods before survey date with a significant decrease of the spline-approximation $\hat{\lambda}_s^a(t)$ and a classification into subperiods according to diagram 4.4. In subperiod 2.6-2.7 yr before survey date $\hat{\lambda}_s^a(t)$ appears to be negative; no definite conclusion is drawn on the course of the real transmission for this period. In subperiod 4.9-5.6 yr before survey date the rate of decrease $\hat{\gamma}_s^a(t)$ of the spline approximation $\hat{\lambda}_s^a(t)$ takes on a value between the lowerbound $\hat{\lambda}(t)$ and the upperbound $\hat{U}(t)$. The estimate of the critical

Table 4.1 Periods before survey date, with a significant decrease in spline trend $\hat{\lambda}_s^a(t)$, classified into subperiods according to diagram 4.4; Mauritius data.

Periods before survey date (yr).

decrease $\hat{\lambda}_s^a(t)$	2.6-5.6	23.6-26.9
($p \leq 0.05$)		
Classification into subperiods		
$\hat{\lambda}_s^a(t) < 0$	2.6-2.7	
$\hat{Y}_s^a(t) < \hat{l}(t)$		
$\hat{l}(t) < \hat{Y}_s^a(t) \leq \hat{u}(t)$	4.9-5.6	
$\hat{Y}_s^a(t) > \hat{u}(t)$	2.8-4.8	23.6-26.9

legenda: $\hat{Y}_s^a(t)$ = rate of change of $\hat{\lambda}_s^a(t)$, $\hat{l}(t)$ = lowerbound, $\hat{u}(t)$ = upperbound.

past-sensitivity $\eta_c(A)$ for ages 4.9-5.6 yr using (4.30) are presented in table 4.2 (see page 143). These values appear to be very low and come close to the estimates $\hat{p}^a(A)$ of the seropositivity rates (linear interpolation). According to diagram 4.4 a past-sensitivity $\eta(A)$ with a value less than the critical past-sensitivity $\eta_c(A)$ would indicate a downward trend of the transmission intensity. The values of $\eta_c(A)$ in this application, however, seem to be too low to allow a subjective indication for a decreasing trend of the transmission intensity in this subperiod.

For the detection of an upward trend in transmission intensity the p-value curve in fig. 4.1 (interrupted line) is used. This curve takes on values ≤ 0.05 in period 6.5-10.1 yr before survey date. If it could be assumed that a) the past-sensitivity is not decreasing for the ages 6.5-10.1 yr

and b) that the past-sensitivity $\eta(A)$ is larger than the critical past-sensitivity $\eta_c(A)$ for these ages, then an upward trend is indicated (see diagram 4.5). The estimates $\hat{\eta}_c(A)$ of the critical past-sensitivity for the ages 6.5-10.1 yr based on (4.22) are presented in table 4.3 (see page 144). If it is supposed that the actual past-sensitivity $\eta(A)$ of the serological test for these ages is larger than $\hat{\eta}_c(A)$ - $\eta(A)$ is larger than 0.21 - then there is an indication for an upward trend of the transmission intensity in period 6.5-10.1 yr before survey date.

Table 4.2 Estimates of the seropositivity rate $p^a(A)$ and the critical past-sensitivity $\eta_c(A)$ for ages 4.9-5.6 yr¹⁾.

age years	seropositivity rate	critical past-sensitivity
	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$
4.9	0.03	0.24
5.0	0.03	0.12
5.1	0.03	0.08
5.2	0.04	0.07
5.3	0.04	0.06
5.4	0.04	0.06
5.5	0.04	0.05
5.6	0.04	0.05

¹⁾critical past-sensitivity $\eta_c(A)$ is defined by (4.26).

Table 4.3 Estimates of the seropositivity rate $\hat{p}^a(A)$ and the critical past-sensitivity $\hat{\eta}_c(A)$ for ages 6.5-10.1 yr¹⁾.

age	seropositivity rate	critical past-sensitivity
years	$\hat{p}(A)$	$\hat{\eta}_c(A)$
6.5	0.06	0.21
7.0	0.07	0.13
7.5	0.07	0.11
8.0	0.08	0.10
8.5	0.08	0.10
9.0	0.08	0.10
9.5	0.09	0.10
10.0	0.09	0.10
10.1	0.09	0.10

¹⁾ critical past-sensitivity $\hat{\eta}_c(A)$ is defined by (4.22).

4.4 EXTENSIONS OF THE MODEL; CORRECTION FOR THE SIMULTANEOUS EFFECT OF THE PAST-SENSITIVITY AND PAST-SPECIFICITY OF THE SEROLOGICAL TEST

In order to assess the simultaneous effect of the past-sensitivity $\eta(A)$ and the past-specificity $\xi(A)$ on the interpretation of the course of the antibody related transmission function $\lambda^a(t)$ two situations are studied:

1. The past-sensitivity $\eta(A)$ as well as the past-specificity $\xi(A)$ are independent of age, are constant and their levels are unknown.
2. The past-sensitivity $\eta(A)$ as well as the past-specificity $\xi(A)$ change with age, their levels, their rates of change and their directions are unknown.

These two situations are direct generalizations of the situations described

in section 4.2.3.3 and section 4.2.3.4 where it was assumed that the past-specificity $\xi(A) = 1$. For situation 1 the exact mathematical relationship between $\lambda^a(t)$ and the real transmission function $\lambda(t)$ has been derived. For situation 2, however, the relationship between the course of $\lambda^a(t)$ and the course of $\lambda(t)$ is derived - as in section 4.2.3.4 - using a linear approximation of the ratio of the seropositivity rate and the cumulative inoculation rate.

Situation 1.

The general relationship between the transmission function $\lambda(t)$ and the antibody related transmission function $\lambda^a(t)$ in a time point A years before survey date is (see Appendix 4)

$$\lambda(A) = \left[\frac{1 - \eta}{\eta - p^a(A)} + 1 \right] \lambda^a(A). \quad (4.31)$$

It is quite surprising to see that formula (4.31) is identical with formula (4.18) found in section 4.2.3.3 where it was assumed that the past-specificity $\xi(A) = 1$. The past-specificity ξ does not appear explicitly as a parameter in the formula. All conclusions drawn in section 4.2.3.3 - especially diagram 4.2 - remain valid if it is additionally assumed that the past-specificity ξ assumes some unknown value. Hence a decreasing antibody related transmission function always refers in situation 1 to a real decrease, irrespective of the actual unknown (constant) levels of the past-sensitivity η and the past-specificity ξ of the serological test.

Situation 2.

The ratio between the seropositivity rate $p^a(A)$ and the cumulative inoculation rate $p(A)$ is called the *seropositivity-inoculation ratio (S-I ratio)*.

and written as $\theta(A)$

$$\theta(A) = \frac{p^a(A)}{p(A)} . \quad (4.32)$$

The S-I ratio is in a sense a generalization of the past-sensitivity $\eta(A)$; from (4.10) it follows that

$$\theta(A) = \eta(A) + [1 - \xi(A)] \frac{1 - p(A)}{p(A)} . \quad (4.33)$$

If the past-specificity $\xi(A) = 1$, then $\theta(A)$ is equal to the past-sensitivity $\eta(A)$ of the serological test. If the past-specificity $\xi(A)$ is not equal to 1 and $p(A)$ is low, then the S-I ratio may assume values larger than 1.

The interpretation of the course of the antibody related transmission function $\lambda^a(t)$, A years before survey date, depends on $p^a(A)$, $\lambda^a(A)$ and

- the value of $\theta(A)$
- its rate of change $\frac{d\theta}{dA}$ and
- its curvature $\frac{d^2\theta}{dA^2}$.

Generally the value, the rate of change as well as the curvature of the S-I ratio are unknown. An analogous procedure is followed as described in section 4.2.3.4: the value $\theta(A)$ and its rate of change are taken into account; the curvature of $\theta(A)$, however, is disregarded. Assuming that the seropositivity rate $p^a(A)$ is increasing with age the following inference is made on $\lambda^a(A)$. If the antibody related transmission function $\lambda^a(t)$ is decreasing in a time point A years before survey date and the rate of decrease $\gamma^a(A)$ is sufficiently large, i.e. larger than a mathematically derived upperbound $u^g(A)$, then the real transmission function $\lambda(t)$ is also decreasing A years before survey date. The generalized upperbound $u^g(A)$ is defined by (see Appendix 4)

$$u^g(A) = \frac{1 + p^a(A)}{2p^a(A)} \lambda^a(A)^2 . \quad (4.34)$$

It is readily seen that the upperbound $u^g(A)$ is larger than the upperbound $u(A)$. The question arises as how close is the upperbound $u^g(A)$ to the upperbound $u(A)$ defined by (4.25). If these bounds come close to each other, then the *maximum linear correction (MLC)* method introduced in section 4.2.4 for the detection of a downward trend of the transmission intensity is rather insensitive for a departure of the assumption 'the past-specificity $\xi(A) = 1$ '. It is easily verified that the ratio between the upperbound $u^g(A)$ and the upperbound $u(A)$, see (4.25) and (4.34), is given by

$$\frac{u^g(A)}{u(A)} = (1 + p^a(A)) (1 - \frac{1}{2}p^a(A)). \quad (4.35)$$

This ratio assumes its maximum value for $p^a(A) = \frac{1}{2}$, this maximum value is $9/8$, thus the upperbound $u^g(A)$ is bounded by $u(A)$ in the following way

$$u(A) \leq u^g(A) \leq 9/8 u(A). \quad (4.36)$$

When it is additionally supposed that the seropositivity rate $p^a(A)$ is smaller than the cumulative inoculation rate, then it can be derived that $u_g(A)$ only assumes values $\leq u(A)$. The inequalities presented in (4.36) imply that the maximum linear correction (MLC) method, which is based on the upperbound $u(A)$, withstands departures from the assumption 'the past-specificity of the serological test is 1'.

4.5 GENERAL DISCUSSION

The methodological aspects of the proposed procedure for the detection of a downward trend in transmission are discussed first. This is followed by comments regarding the application of the model. Step 1 of the procedure (see section 4.2.4) is based on statistical inference concerning the spline-trend of the antibody related transmission function. Step 2 is

based on the maximum linear correction (MLC) method for the factor of past-sensitivity. The statistical aspects of spline-trend estimation and testing have already been discussed in chapter 2. Here the methodological aspects of the MLC method are discussed such as:

- a) ignoring the past-specificity of the serological test
- b) the transfer of formula (4.27) which only concerns the relationship between the two "intensity" functions $\lambda^a(t)$ and $\lambda_s(t)$, to their associated spline approximations $\hat{\lambda}_s^a(t)$ and $\lambda_s(t)$
- c) disregarding the curvature of the past-sensitivity $\eta(A)$.

4.5.1 The past-specificity of the test

The procedure for the detection of a downward trend in transmission intensity uses the spline approximation $\hat{\lambda}_s^a(t)$ as an estimate of the antibody related transmission function $\lambda^a(t)$. This spline approximation $\hat{\lambda}_s^a(t)$ was defined in chapter two as the first derivative of a natural cubic spline which passes through the origin $(0,0)$ and the points $(A_j, -e \log(1 - \hat{p}^a(A_j)))$ where $j=1,\dots,k$. This implies that the seropositivity rate for age zero is implicitly considered to be zero. Taking into account a past-specificity ξ would mean that the seropositivity rate for age zero has to be considered as $1 - \xi$ and consequently the natural cubic spline should pass through $(0, -e \log \xi)$ instead of through the origin $(0,0)$. Generally, however, the past-specificity of the serological test can be made to come close to 1 by taking the discriminative titre level sufficiently high. In this situation the difference between the natural cubic spline approximation which passes through $(0,0)$ and that which passes through $(0, -e \log \xi)$ might be disregarded. Anyway this difference will diminish rapidly for time points t situated further in the past; it may already be ignored for time points t

after the third knot T_2 before survey date (T_2 is the midpoint of age group A_2).

Highly specific test methods are required especially in areas where the prevalence of antibodies is low. One should try to obtain a past-specificity of about 0.99 for seropositivity rates $p^a(A) < 0.10$. When this can not be obtained one should be extremely cautious in interpreting the spline approximation $\lambda_s^a(t)$ in the corresponding period before survey date. In fact if the past-specificity ξ is less than 0.95, the seropositivity rate $p^a(A) < 0.10$ and the past-sensitivity $\eta(A) > 0.50$, then it can be shown that the seropositivity rate $p^a(A)$ is actually larger than the true cumulative inoculation rate $p(A)$. This will result in an overestimation of the level of transmission in the corresponding period near the survey date.

In section 4.4 it was shown that a constant past-sensitivity and a constant past-specificity do not disturb the interpretation of a decreasing antibody related transmission function. The rate of decrease in $\lambda^a(t)$ is in the case of a constant past-sensitivity and a constant past-specificity even smaller than the rate of decrease in the real transmission function $\lambda(t)$. Moreover the MLC method for the factor of past-sensitivity seems rather insensitive for the additional incorporation of the factor of past-specificity. Diagram 4.2 remains completely valid and furthermore the upperbound $u(A)$ used in the MLC method is only slightly affected if the simultaneous effect of an age related past-sensitivity $\eta(A)$ and past-specificity $\xi(A)$ is taken into account (see (4.36)).

4.5.2 Relationship between the course of the spline approximations $\lambda_s^a(t)$ and $\lambda_s(t)$

The MLC method was based on the relationship between the rate of decrease $\gamma^a(A)$ of the antibody related transmission function and the rate of decrease

$\gamma(A)$ of the real transmission function (see formula (4.27));

if $\gamma^a(A) > u(A)$ then $\gamma(A) > 0$,

where the upperbound $u(A)$ was defined by (see (4.25))

$$u(A) = \frac{\lambda^a(A)^2}{p^a(A)(2 - p^a(A))} .$$

This relationship between $\gamma^a(A)$ and $\gamma(A)$, which is valid if the curvature of the past-sensitivity $\eta(A)$ is disregarded, has been used as a guide to develop the MLC method for the factor of past-sensitivity. The question arises as to whether there is an analogous relationship between the spline approximations of $\gamma^a(A)$ and $\gamma(A)$. In appendix 4 it is shown that, under certain mathematical conditions and appropriate definitions of spline approximations $p_s(A)$, $p_s^a(A)$ and $\eta_s(A)$ of $p(A)$, $p^a(A)$ and $\eta(A)$ respectively, the following relationship holds

$$\text{if } \gamma_s^a(A) > u_s(A) \text{ then } \gamma_s(A) > 0, \quad (4.37)$$

where the upperbound $u_s(A)$ is defined by

$$u_s(A) = \frac{\lambda_s^a(A)^2}{p_s^a(A)(2 - p_s^a(A))} . \quad (4.38)$$

This relationship between $\gamma_s^a(A)$ and $\gamma_s(A)$ is valid if the curvature of the spline approximation $\eta_s(A)$ of the past-sensitivity is disregarded. The spline approximation $p_s^a(A)$ in the formula for $u_s(A)$ is defined by

$$p_s^a(A) = 1 - \exp\left(-\int_0^A \lambda_s^a(t) dt\right). \quad (4.39)$$

The implication of the above is that an estimator based on (4.39) has to be preferred to the simple linear interpolation technique that has been used for the estimation of $p^a(A)$ in the formula for $u(A)$.

4.5.3 The curvature of the past-sensitivity

The value $r(A)$, the rate of change $\frac{dn}{dA}$ and the curvature $\frac{d^2n}{dA^2}$ of the past-sensitivity depend on age and are unknown. When it is not acceptable to make any assumption with respect to $n(A)$, $\frac{dn}{dA}$ and $\frac{d^2n}{dA^2}$, then correction for the factor of past-sensitivity is impossible and consequently the serological profile of the community cannot give information on the course of the transmission intensity in that community. It seems methodologically acceptable, however, to use an indirect correction method. The reliability of the *maximum linear correction (MLC) method* in this study is not based on statistical inference but on an indirect structural argument, i.e. if the rate of decrease of the antibody related transmission function cannot be explained by *any* combination of value and rate of change of the past-sensitivity then the decision is made that the data provide evidence for a real decrease in the transmission intensity.

What is the effect of disregarding the curvature of the past-sensitivity in using the MLC method? When this method is applied then a concave curvature of the past-sensitivity cannot overturn the decision that a real decrease in transmission intensity has occurred. However a convex curvature of the past-sensitivity with age may undo the decision. This can be a potential source of errors if the MLC method is used. Further studies will be needed to explore the possibility that a "large" convex change of the past-sensitivity $n(A)$ with age occurs simultaneously with an increase in the transmission intensity.

If more information becomes available on the factor of past-sensitivity then it is preferable to develop a direct correction method based on estimates of the past-sensitivity $n(A)$ of the serological test. Estimates of the cumulative inoculation rate $p(A)$ would follow for different age groups and a direct assessment of the trend of the transmission intensity would then be possible using the time-dependent transmission model described in chapter 2.

4.5.4 The validity of the two-step procedure

The validity of the two-step procedure cannot be expressed in terms of error probability. Only the first step of the procedure is based on statistical inference. A complete stochastic approach would also take into account the statistical fluctuations in the second step of the procedure. This would mean the development of a statistical test for the following hypothesis:

$$H_0: \gamma_s^a(A) - u_s(A) \leq 0 \text{ against } H_1: \gamma_s^a(A) - u_s(A) > 0.$$

This is not taken into consideration in this phase of the study for reasons of complexity of the distribution of the statistic $\hat{\gamma}_s^a(A) - \hat{u}_s(A)$. This, however, may be the object of further research.

The validity of the two-step procedure should be evaluated by applying the procedure in areas which are in different phases of malaria control. In order to define its role in anti-malaria campaigns the results of the analysis have to be compared with parasitological and/or entomological data obtained from malaria surveillance activities.

The methodology in this chapter has been based on the assumption that the population under study is homogeneous. Here the question arises as to how one should adapt the procedure to detect a downward trend in transmission in a non-homogeneous population. The two-step procedure can be generalized using the concept "average antibody related transmission function". This function, written as $\Lambda^a(t)$, is defined in an analogous way as the antibody related transmission function $\lambda^a(t)$ i.e. $\Lambda^a(t)$ is the "transmission function" based on the average¹⁾ seropositivity rate $P^a(A)$. The value of $\Lambda(t)$ and $\Lambda^a(t)$ respectively in a time point A years before survey date is then equal to (see also 4.9)

¹⁾ The average seropositivity rate $P^a(A)$ is defined in an analogous way as the average cumulative inoculation rate $P(A)$, see chapter 3 formula (3.10).

$$\Lambda(A) = \frac{\frac{dP(A)}{dA}}{1 - P(A)} \quad \Lambda^a(A) = \frac{\frac{dP^a(A)}{dA}}{1 - P^a(A)}, \quad (4.40)$$

where $P(A)$ = average cumulative inoculation rate,

and $P^a(A)$ = average seropositivity rate.

Using the concept *average past-sensitivity*, written as $H(A)$, the relationship between the rate of change of $\Lambda^a(t)$ and the rate of change of the true average transmission function $\Lambda(t)$ can be derived. Assuming that the past-specificity $\xi(A) = 1$ in all subpopulations considered, the average past-sensitivity $H(A)$ for a non-homogeneous population has been formally defined as the ratio of $P^a(A)$ and $P(A)$,

$$\text{average past-sensitivity } H(A) = \frac{P^a(A)}{P(A)}. \quad (4.41)$$

If the curvature of $H(A)$ is disregarded then an analogous relation holds as has been derived for a homogeneous population (see 4.27) i.e.,

$$\text{if } \Gamma^a(A) > U(A) \text{ then } \Gamma(A) > 0, \quad (4.42)$$

$$\text{where } U(A) \text{ is defined by } U(A) = \frac{\Lambda^a(A)^2}{P^a(A)(2 - P^a(A))}. \quad (4.43)$$

Furthermore the relationship between the spline approximations $\Gamma_s^a(A)$ and $\Gamma_s(A)$ is analogous to the relationship between $\gamma_s^a(A)$ and $\gamma_s(A)$ (see 4.37);

$$\text{if } \Gamma_s^a(A) > U_s(A) \text{ then } \Gamma_s(A) > 0, \quad (4.44)$$

$$\text{where } U_s(A) = \frac{\Lambda_s^a(A)^2}{P_s^a(A)(2 - P_s^a(A))} \quad (4.45)$$

$$\text{and } P_s^a(A) = 1 - \exp\left(- \int_0^A \Lambda_s^a(t) dt\right). \quad (4.46)$$

This relationship between $\hat{\Gamma}_s^a(A)$ and $\Gamma_s(A)$ is valid if the curvature of the spline approximation $H_s(A)$ of the average past-sensitivity is ignored (for further details see Appendix 4).

The procedure for the detection of a downward trend of the transmission intensity in a non-homogeneous population could be based on (4.44). The analogy of step 1 of the two-step procedure for a homogeneous population would be the determination of periods before survey date in which the rate of change $\hat{\Gamma}_s^a(t)$ of the average spline-trend $\hat{\Lambda}_s^a(t)$ is statistically significant (trend-change test, $p \leq 0.05$ (one-sided), see chapter 3). The analogon for step 2 would follow from (4.44) using estimates of the parameters involved in this formula. In chapter 5 this method will be used to detect a downward trend in transmission in the Kilinochchi district in Sri Lanka. When the population is actually homogeneous then (4.44), (4.45) and (4.46) are equivalent with (4.37), (4.38) and (4.39) respectively. Therefore the two-step procedure suggested for a non-homogeneous population can be considered as a generalization of the two-step procedure proposed for a homogeneous population (section 4.2.4).

Another possibility for further analysis of periods in which the overall trend $\hat{\Lambda}_s^a(t)$ is decreasing might be to select for each time point A of these periods all those sub-areas which show a decreasing spline approximation $\hat{\lambda}_s^a(t)$ and fulfil the condition $\hat{\gamma}_s^a(A) > \hat{u}_s(A)$. These sub-areas might then be considered to have a downward trend of transmission intensity A years before survey date.

4.5.5 The critical past-sensitivity

The critical past-sensitivity $\eta_c(A)$ is a mathematically defined function. It is derived following the assumption that the curvature of the past-

sensitivity $\eta(A)$ can be disregarded. This function could be useful for the interpretation of the rate of change ($\gamma^a(A)$) of the antibody related transmission function in a time point A years before survey date. For $\gamma^a(A) < 0$ the critical past-sensitivity is defined by (4.22); for $\ell(A) < \gamma^a(A) \leq u(A)$ it is defined by (4.26).

a) *antibody related transmission function is increasing ($\gamma^a(A) < 0$)*

With regard to the detection of a real increase in transmission intensity the critical past-sensitivity $\eta_c(A)$ is of little practical value, unless it could be assumed that $\eta(A)$ is not decreasing with age (see diagram 4.5). An increase in $\lambda^a(t)$, A years before survey date is then interpreted as a real increase if the actual unknown past-sensitivity $\eta(A)$ can be assumed to be larger than the critical past-sensitivity $\eta_c(A)$. For the youngest children certainly an age interval will exist in which the past-sensitivity $\eta(A)$ is decreasing with age. Any type of decreasing past-sensitivity $\eta(A)$, i.e. linear, concave or convex may explain an increase in the antibody related transmission function. Therefore an increase in $\lambda^a(t)$ in the corresponding period before survey date might be actually the result of the disturbing effect of the factor of past-sensitivity. Thus caution is needed in interpreting an increase in the antibody related transmission function, especially if the period of increase is near to the survey date.

b) *antibody related transmission function is decreasing ($\gamma^a(A) > 0$)*

If the past-sensitivity $\eta(A)$ is independent of age or decreasing with age, then the critical past-sensitivity $\eta_c(A)$ is not involved in the interpretation of $\lambda^a(t)$; a decrease in $\lambda^a(t)$ then reflects a real decrease (see diagram 4.5). Only if $\eta(A)$ increases with age or its direction of change is unknown $\eta_c(A)$ may be of use. If in this situation the actual value of the past-sensitivity $\eta(A)$ is assumed to be less than $\eta_c(A)$, then the decrease

in $\lambda^a(t)$ is still considered to reflect a real decrease in transmission intensity. Clearly the practical value of the concept critical past-sensitivity $\eta_c(A)$ for the detection of a downward trend of the transmission intensity is rather limited. Although the critical past-sensitivity can be estimated from the serological data, the decision $\eta(A) < \eta_c(A)$ or $\eta(A) > \eta_c(A)$ remains subjective. Only when the critical past-sensitivity $r_c(A)$ takes on values near to 1, then it can be decided that the actual past-sensitivity $\eta(A)$ is smaller than $\eta_c(A)$.

The critical past-sensitivity $\eta_c(A)$ as well as the unknown past-sensitivity $\eta(A)$ are functions of age which assume values between the seropositivity rate $p^a(A)$ and one. For a very large seropositivity rate - $p^a(A)$ approaching one - it will hardly be possible to decide whether $\eta(A) < \eta_c(A)$ or $\eta(A) > \eta_c(A)$. Hence the critical past-sensitivity will not be of practical value for the older age-groups in highly endemic areas. Furthermore when the seropositivity rate $p^a(A)$ approaches one then the upperbound $u(A)$ moves towards the lowerbound $\ell(A)$; this also indicates that the practical value of the critical past-sensitivity will disappear for the larger seropositivity rates.

4.5.6 Comments on the application

In Mauritius a real decrease in the trend of the transmission intensity is indicated for two periods before the survey date. This decrease occupies the entire duration of the earlier period as already indicated by application of the time-dependent transmission model. For the more recent period of decrease of $\hat{\lambda}_s^a(t)$ it applies only to a sub-period. With regard to the period of increase, 6.5-10.1 yr before the survey date, this increase does not allow interpretation unless it could be assumed that the past-sensitivity $\eta(A)$

of the serological test is not decreasing for these ages (see diagram 4.5).

There is no information available on the course of the past-sensitivity.

Therefore the course of the real trend of the transmission intensity in period 6.5-10.1 yr remains uncertain. The unexplained increase in the spline approximation $\hat{\lambda}_s^a(t)$ (chapter 2) might be an artefact caused by an age-related decreasing past-sensitivity $\eta(A)$ for children of 6-10 yrs old. It can be concluded that the methodology for the detection of a downward trend of the transmission intensity produces results which appear to fit quite well with the history of malaria in Mauritius as described in chapter 2, section 2.4.7.

4.6 SUMMARY

A procedure is proposed for the detection of a downward trend in transmission intensity using the age related rising seropositivity rates in a homogeneous population. The procedure consists of two steps. Step 1 is based on statistical inference using the time-dependent transmission model developed in chapter 2. In step 2 a correction for the factor of past-sensitivity of the serological test is performed. This correction, called *maximal linear correction (MLC)*, takes into account the unknown value and the unknown rate of change of the age-related past-sensitivity.

Sero-epidemiological data from Mauritius, already analysed in chapter 2 (step 1 of the procedure), are used to illustrate the application of the procedure. The results fit quite well with the history of malaria in Mauritius.

APPLICATIONS

5

5.1 GENERAL INTRODUCTION

The previous chapters mainly concerned the development of models for the detection of change in the trend of the transmission intensity. Using the time-dependent transmission model developed in chapter 2 and the maximum linear correction (MLC) method for the factor past-sensitivity introduced in chapter 4, a two step procedure has been proposed in section 4.2.4 for the detection of a downward trend of the transmission intensity in a homogeneous population. This procedure was extended in section 4.5.4 in order to apply it also to non-homogeneous populations. The validity of these procedures should be evaluated by their application to data from the field. In this chapter serological data from Guyana, Surinam, Sri Lanka, Nigeria and Panama are (re-)analysed in order to establish periods in which a decrease in the trend of the transmission has occurred. The results are compared with the

available data on the history of malaria transmission in these study areas.

The Mauritius data have been used in section 4.3 to illustrate the application of the two-step procedure. The type of presentation used in that section will be the standard for the applications of the procedure in this chapter.

5.2 SERO-EPIDEMOIOLOGICAL INVESTIGATIONS OF MALARIA IN GUYANA

5.2.1 *Introduction*

Lobel et al. (1976) have reported on the results of a two-stage cluster survey carried out in 1973 and a partial follow-up and resurvey one year later in the interior of Guyana where the transmission of malaria is considered to be interrupted. The specific objectives were firstly to determine the serological indices in relation to the parasitologic data as well as the malaria surveillance information and secondly to assess whether the serological data could support the hypothesis that there had been little or no malaria transmission in the interior since 1968. The purposes of this re-analysis are:

- the determination of periods with a downward trend of transmission intensity using the two-step procedure described in section 4.2.4
- the comparison of the results of this analysis with those reported by *Lobel et al.*, who have used the constant infection rate model of *Draper et al.* (1972) for the analysis of the data of the 1973 survey.

5.2.2 *The design of the study*

A brief summary is given of the methods and materials used in the Guyana study as reported by *Lobel et al. (1976)*. Four sectors in the interior of

Guyana were selected in which malaria case detection operations had been carried out irregularly between 1968 and 1971 and not at all in 1972; in these areas malaria transmission could easily have escaped detection. The estimated population size was 5,560 (1970 census) and consisted of 155 villages in the four survey sectors. A total of 45 villages was included in the survey and about 1,000 persons were examined to estimate the serological status in the age groups 1-4, 5-9, 10-19, 20-29, 30-39 and 40-59 years. A resurvey took place one year later, in May 1974, and was carried out in the 10 localities in which the sera of one or more children born after 1968, had had a positive reactivity in the first survey. This resurvey included all these children, their household members and neighbours and also additional children born after 1968. Blood specimens and detailed histories were obtained from 352 persons, 147 of whom had been included in the 1973 survey.

In both surveys the indirect haemagglutination (IHA) test for malaria was carried out with *P.falciparum* and *P.vivax* antigens. The blood samples for serological examination were collected on filter paper. A reaction was considered to be positive when agglutination occurred in the first dilution with either antigen or both. This was considered to correspond to a serum titre of 32. For further details see *Lobel et al.* (1976).

5.2.3 Results

Table 5.2.1 (see page 162) presents the prevalence of antibodies by age in the interior of Guyana. These data were analysed using the constant infection rate model of *Draper et al.* (1972). Graphic representation of the relationship between age and sero-negativity rate in persons above and below 20 years old on a log scale showed two straight lines with different slopes, which suggested that the probability of being infected in a year (*R*) had

Table 5.2.1 Results of the IHA test carried out on serum samples collected in the interior of Guyana in 1973 (positive is titre $\geq 1:32$).

Age group (in years)	number exam.	number positive	%	\hat{R}
1-4	155	1	0.6	
5-9	158	1	0.6	0.06%
10-19	189	2	1.1	
20-29	109	7	6.4	
30-39	126	33	26.2	0.91%
40-59	198	86	43.4	

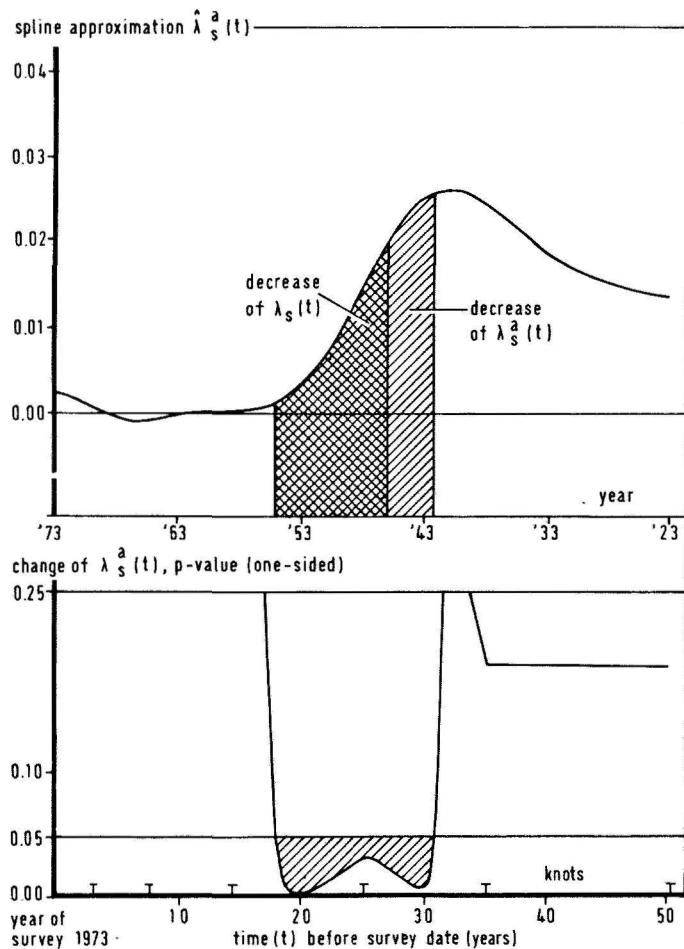
Source: Sero-epidemiologic investigations of malaria in Guyana, Lohel et al. (1970)

\hat{R} = estimated probability of being infected in a year, method Draper et al., 1972; constant infection rate model.

been fairly constant but of differing intensity in each of the two population segments. The estimated R-values in each of the two population segments were 0.06% and 0.91% respectively (Lohel et al. 1970).

The results of the analysis of table 5.2.1 using the time-dependent transmission model with MLC correction for the factor past-sensitivity are shown in fig. 5.2.1. A decrease in the spline approximation $\hat{\lambda}_s^a(t)$ of the antibody related transmission function is found in period 17.8-30.6 yr before survey date ($p \leq 0.05$, one-sided test, lower-half of the figure). Correction for the factor past-sensitivity, step 2 of the procedure, shows a decrease in the spline approximation $\lambda_s(t)$ of the real transmission function, in the sub-period 17.8-26.9 yr before survey date (upper-half of fig. 5.2.1). Since the survey took place in May 1973, the methodology applied indicates a downward

Figure 5.2.1 Application of the two-step procedure for the detection of a downward trend of the transmission intensity in Guyana.



See general legenda page 247:

$\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

$\lambda_s(t)$ = spline-trend of the transmission intensity

In the subperiod 27.0-30.4 yr before survey date the rate of decrease $\hat{\gamma}_s^a(t)$ of the spline approximation $\hat{\lambda}_s^a(t)$ takes on a value between the lower-bound $\hat{\lambda}(t)$ and the upperbound $\hat{U}(t)$. The estimates of the critical past-sensitivity $\hat{n}_c^a(A)$ for ages 27.0-30.4 are presented in table 5.2.2. If the actual past-sensitivity for these ages would be smaller than the critical past-sensitivity $\hat{n}_c^a(A)$, then there would be an indication for a real decrease in the trend of transmission in this subperiod. Because of the rapid decline of the critical past-sensitivity in age interval 27.0-30.4 yr, only a very small period about 27 years before survey date might be additionally considered for a real decrease in the trend of transmission intensity. In the remaining very small subperiod 30.5-30.6 yr before survey date, the course of the spline trend of the real transmission intensity is uncertain due to

Table 5.2.2 Guyana; estimates of the seropositivity rate $\hat{p}^a(A)$ and the critical past-sensitivity $\hat{n}_c^a(A)$ for ages 27.0-30.4 yr.

age years	seropositivity rate $\hat{p}^a(A)$	critical past-sensitivity $\hat{n}_c^a(A)$
27.0	0.10	0.77
27.5	0.11	0.33
28.0	0.12	0.24
28.5	0.13	0.21
29.0	0.14	0.19
29.5	0.15	0.18
30.0	0.16	0.18
30.4	0.17	0.17

the fact that the rate of decrease $\hat{\gamma}_s^a(t)$ is smaller than the lowerbound $\hat{\lambda}(t)$. (cf. chapter 4, diagram 4.4).

5.2.4 Discussion

The results of the application of the two-step procedure to the serological data of the 1973 survey show a remarkable resemblance with the malaria history of the coast and near interior areas in Guyana. To substantiate this the main facts of the malaria history in Guyana as reported by Lobel *et al.* (1976) are summarized here.

Malaria used to be wide spread throughout the country with *A.darlingi* and *A.aquasalis* as the main vectors of malaria parasites. An extensive DDT house spraying campaign was carried out in the coastland and near interior from 1947 to 1950. In subsequent years, general insecticide spraying was limited to the upper estuaries until it was discontinued entirely in 1958. The DDT spraying resulted in the elimination of *A.darlingi* and endemic malaria from the coastal and near interior regions by 1950 (Giglioli, 1951); this part of Guyana was therefore placed in the maintenance phase at the inception of the malaria eradication programme in 1960. Since that time, a focal outbreak occurred in 1961 near Georgetown with *A.aquasalis* as the vector and only 15 other indigenous cases of malaria were detected in the maintenance phase area.

In the remainder of Guyana, which contrasts sharply with the densely populated coastal areas, only limited DDT spraying was carried out before 1960. This part of the country, which is sparsely populated and covered by a thick jungle (total population 46,000), was then placed in the attack phase and the antimalaria measures included DDT house spraying, distribution of chloroquinized salt and active and passive case detection. The main vector was sylvan *A.darlingi*. Since 1960 outbreaks have been detected in the north-

west region in 1966, the Cuyuni-Mazaruni region in 1966-1967 and the Rupununi region in 1972. In 1970 the attack phase area was advanced into the consolidation phase. The malaria surveillance had not detected any indigenous cases of malaria since 1968 except in some areas along the Brazilian border, in the Rupununi region, but complete and regular malaria surveillance in the entire country had been difficult due to logistical and operational limitations.

The analysis of the Guyana data using the two-step procedure indicates a downward trend of the transmission intensity in the years 1946-1955 in four sectors in the interior of Guyana. The new model - which is not based on an a posteriori classification of age groups in two population segments and which takes into account a variable inoculation rate and the factor past-sensitivity gives a more detailed picture of the trend of the transmission intensity than the constant infection rate model of Draper *et al.* (1972). The striking difference between the R-values in the two population segments (0.06% and 0.91%, table 5.2.1) was related to the observation that persons under 20 years old were born after the completion of the successful eradication campaign. This campaign resulted in the elimination of malaria from the coastal and near interior areas where the disease had been highly endemic previously.

The results of the present analysis confirm the interpretation of Lobel *et al.* (1976) that the transmission in the interior had been dependent on the level of malaria endemicity on the coast. This epidemiological dependence was explained by the population mobility between the coast and the interior localities. The scattering of the population groups within the interior might have contributed to the regional outbreaks in that part of the country.

In the interior a special antimalaria campaign was not undertaken until 1961-1970. The spline approximation $\hat{\lambda}_s^a(t)$ of the antibody related transmission function, however, does not provide evidence for a downward trend of the

transmission intensity after 1960. *Lobel et al.* (1976) suggest that the disappearance of malaria from the interior is more related to the elimination of malaria transmission on the coast than to the specific malaria campaign in the interior. It cannot be excluded, however, that a relatively rapid fading of antibodies in the individuals born after 1960 might have affected the ability of the procedure to detect a downward trend of the transmission intensity in that period.

Past-specificity of the serological test

Lobel et al. (1976) report a low frequency of nonspecific agglutination; only two out of 190 sera (1.1%) showed agglutination in dilution 1:16 after absorption with test cells sensitized with "control" antigen. In order to satisfy the requirement of a high specificity the discriminative titre level was chosen as 1:32. Nonspecific agglutination was not observed at this dilution, which indirectly indicates that the past-specificity of the serological test in the Guyana survey was close to 100%.

As already expected from the theoretical considerations in chapter 4 (section 4.4), the use of the upperbound $u^g(A)$ instead of $u(A)$ yields practically the same results; a downward trend in transmission intensity is indicated in period 17.8-26.7 yr before survey date. Hence if $u^g(A)$ is used only a very small period 26.8-26.9 yr before survey date is lost in which a downward trend in transmission is no longer indicated. This finding underlines the statement in section 4.5.1 on the robustness of the MLC method with respect to disregarding the factor past-specificity.

Recent transmission

No malaria parasites could be found in any of the blood slides even

though concentrated blood smears (*Bawling et al.* 1966) were made during the follow-up survey in 1974. Absence of two fold or higher titre increases in the follow-up of 147 persons and confirmation that 14 children born after 1968 with a titre \geq 16 had no history indicative of malaria infection or of travel outside the area support the conclusion that there has been little or no malaria transmission in the interior since 1968.

The re-analysis of the Guyana data gives support to the idea that the time-dependent transmission model with MLC for the factor past-sensitivity can be used as a tool for the study of the epidemiology of malaria, especially in areas where the incidence of malaria is too low to be evaluated reliably by blood slide examination alone.

5.3 MALARIA IN SURINAM, A SERO-EPIDEMIOLOGICAL STUDY

5.3.1 *Introduction*

Van der Kaay (1975) has analysed the results of the indirect haemagglutination test (IHA) at village level during an evaluation of malaria control activities in Surinam. Two cross-sectional surveys were carried out with an interval of one year, during August and September 1973 and the same period of 1974. Villages were selected in non-malarious areas and in areas with endemic or epidemic malaria. The constant infection rate model was applied to estimate the probability of a person becoming infected with malaria in one year (R-value). The field study was carried out in 4 areas. At least for two areas the model assumption of a 'constant probability of being infected' is not fulfilled. The conclusion therefore was that it was necessary to revise the constant infection rate model of *Draper, Voller and Carpenter* (1972).

This study reports the results of the application of the time-dependent

transmission model with maximal linear correction for the factor past-sensitivity. The specific objectives were firstly to investigate whether the two-step procedure, described in section 4.2.4, applied to the 1973 and 1974 data produces consistent results and secondly to assess whether these results are in line with the history of malaria in the localities where the field study was carried out.

5.3.2 Results of the analysis using the constant infection rate model

The four areas included in the survey are situated along the coast, the Upper Surinam River, Stoelmans Island and in the Central Jungle Area. The serological data as reported by van der Kaay (1970) are presented in table 5.3.1 (see page 170). Estimates of the infection probability R and its standard error (σ), based on three age groups between the age of 1-10 years, using the maximum likelihood (ML) and the least squares (LS) methods are presented in table 5.3.2 (see page 171). The model assumption of 'a constant probability of being infected' is not fulfilled at least for Stoelmans Island and the Central Jungle Area. Thus it was not surprising that a Chi-square test for goodness of fit, carried out on the data, strongly rejected the hypothesis that in the case of these two areas the data fit the constant infection rate model.

5.3.3 Application of the two-step procedure

No attempt has been made to apply the two-step procedure to the data from the Coastal Area; these data do not fulfil the assumptions required for the application (see discussion). The methodology applied to the data from Upper Surinam River and Stoelmans Island respectively does not provide evidence for a downward trend of the transmission intensity.

Table 5.3.1 Results of the IHA test for malaria carried out on serum samples collected in 1973 and 1974 in four areas of Surinam (*P.falciparum* antigen, positive is titre $\geq 1:40$).

Age group	Area and year of survey											
	Coastal Area			Upper Surinam River			Stoelmans Island			Central Jungle Area		
	no exam.	no pos.	% pos.	no exam.	no pos.	% pos.	no exam.	no pos.	% pos.	no exam.	no pos.	% pos.
1973												
12-23 mo	10	1	10	9	1	11	11	4	36	12	1	8
2-4 yr	22	1	5	28	12	43	49	15	31	30	12	40
5-9 yr	25	1	4	26	18	70	60	25	42	44	18	41
10-19 yr	47	1	2	25	21	84	48	34	71	66	61	92
20-39 yr	23	2	9	50	45	90	84	77	92	62	62	100
40-59 yr	16	0	0	16	15	94	32	29	91	31	31	100
1974												
12-23 mo	9	0	0	8	1	13	17	2	12	15	3	20
2-4 yr	30	0	0	38	14	37	42	7	17	37	9	24
5-9 yr	53	0	0	30	20	67	58	12	21	43	11	26
10-19 yr	65	3	5	17	12	71	50	22	44	74	57	77
20-39 yr	31	0	0	60	51	85	63	52	83	72	70	97
40-59 yr	25	1	4	26	24	92	20	18	90	35	35	100

Source: van der Kaay (1975). Malaria in Surinam, a sero-epidemiological study. Meppel, Krips B.V. (thesis).

The areas refer to inhabitants examined in:

Totness and Friendship (Coastal Area)

Dang and Kambaloea (Upper Surinam River)

area around Stoelmans Island (Stoelmans Island)

Alalaparoe (Central Jungle Area).

In these two areas, the one-sided p-value function for the detection of a decrease in the spline trend $\lambda_s^a(t)$ does not fall below the critical value 0.05.

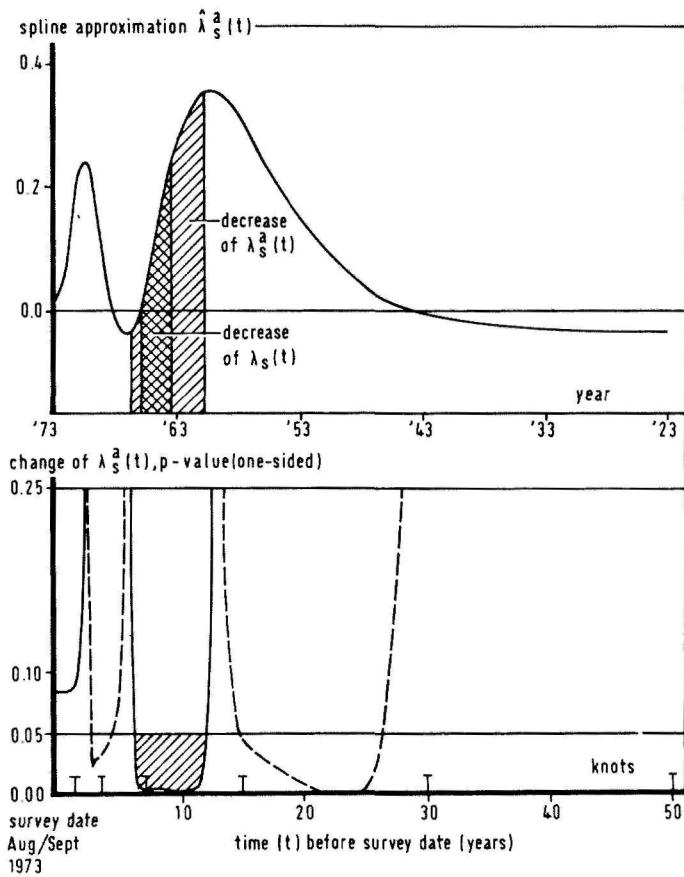
The analysis of the data from the Central Jungle Area yields in both surveys statistically significant results in step 1 of the procedure. The estimation of the spline trend $\lambda_s^a(t)$ and the one-sided p-value function for the detection of periods of change in $\lambda_s^a(t)$ are presented in fig. 5.3.1 (1973 survey) and fig. 5.3.2 (1974 survey) (see pages 172 and 173).

Table 5.3.2 Estimates of the infection probability R and its standard error (σ), based on the seropositivity rates in three age groups between the age of 1-10 years, using the maximum likelihood and the least squares method.

<i>Method of estimation</i>	Area	Year	<i>Maximum likelihood</i>		<i>Least squares</i>	
			\hat{R}	$\hat{\sigma}$	\hat{R}	$\hat{\sigma}$
Coastal Area		1973	1.1%	\pm 0.6%	0.9%	\pm 0.5%
		1974	0.0%	\pm -	0.0%	\pm -
Upper Surinam River		1973	14.2%	\pm 2.8%	14.4%	\pm 3.2%
		1974	12.8%	\pm 2.4%	13.2%	\pm 2.8%
Stoelmans Island		1973	8.4%	\pm 1.3%	8.1%	\pm 1.2%
		1974	3.8%	\pm 0.8%	3.6%	\pm 0.8%
Central Jungle Area		1973	8.3%	\pm 1.6%	8.0%	\pm 1.5%
		1974	5.4%	\pm 1.2%	4.9%	\pm 1.1%

Source: *van der Kaay (1976). Malaria in Surinam, a sero-epidemiological study.*

Figure 5.3.1 Application of the two-step procedure for the detection of a downward trend of the transmission intensity in Alalaparoe, Central Jungle Area, Surinam (survey 1973).

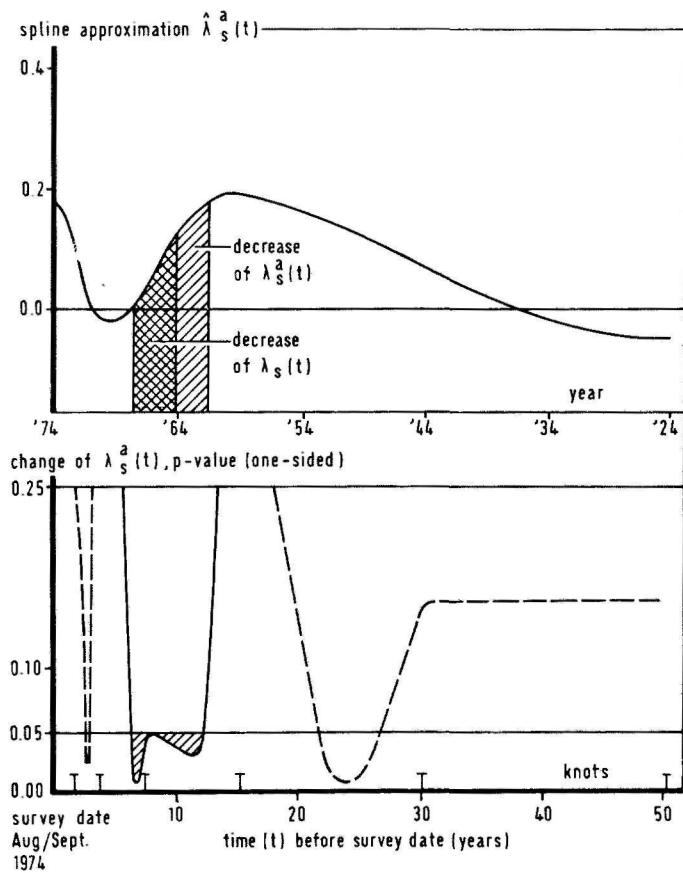


See general legenda page 247:

$\lambda_s^\alpha(t)$ = spline-trend of the antibody related transmission function

$\lambda_s(t)$ = spline-trend of the transmission intensity

Figure 5.3.2 Application of the two-step procedure for the detection of a downward trend of the transmission intensity in Alalaparoe, Central Jungle Area, Surinam (survey 1974).



See general legenda page 247:

$\hat{\lambda}_s^a(t)$ = spline-trend of the antibody related transmission function
 $\lambda_s^a(t)$ = spline-trend of the transmission intensity

The 1973 survey as well as the 1974 survey indicate 3 periods of change in $\lambda_s^a(t)$ and are presented in table 5.3.3. The periods of decrease i.e. 6.4-12.1 yr before survey date (survey 1973) and 6.2-12.2 yr before survey date (survey 1974) are further analysed in step 2 of the procedure. Application of the MLC method for the factor past-sensitivity yields subperiods in which a decrease in the spline trend $\lambda_s^a(t)$ of the real transmission intensity is indicated (see the double shaded area in fig. 5.3.1 and fig. 5.3.2). The 1973 survey provides evidence for a decrease in $\lambda_s^a(t)$ in period 7.1-9.6 yr before survey date, the 1974 survey indicates a decrease in $\lambda_s^a(t)$ in period 6.2-9.7 yr before survey date.

Central Jungle Area, critical past-sensitivity

In the 1973 as well as in the 1974 survey , a period - 9.7-9.8 yr and 9.8-10.4 yr before the survey date respectively - is found in which $\hat{\lambda}_s^a(t)$ is decreasing ($p \leq 0.05$) and in which the rate of change $\hat{\gamma}_s^a(t)$ takes on a value between the lowerbound $\hat{\lambda}(t)$ and the upperbound $\hat{U}(t)$. The estimates

Table 5.3.3 Periods of change of the spline trend $\hat{\lambda}_s^a(t)$ of the antibody related transmission function ($p \leq 0.05$; one-sided). Central Jungle Area, Alalaparoe.

Periods before survey date (yr)		
change	survey 1973	survey 1974
decrease	6.4 - 12.1	6.2 - 12.2
increase	2.8 - 4.5	2.7 - 2.9
increase	14.8 - 26.4	21.5 - 26.7

of the critical past-sensitivity $\hat{\eta}_c(A)$ in corresponding age intervals 9.7-9.8 yr (survey 1973) and 9.8-10.4 yr (survey 1974) are presented in table 5.3.4. The critical past-sensitivity associated with the periods of increase in the spline trend $\hat{\lambda}_s^a(t)$ are presented in table 5.3.5 (see page 176).

The interpretation of the values of the critical past-sensitivity $\hat{\eta}_c(A)$ depends on the subjective comparison of $\eta(A)$ with $\hat{\eta}_c(A)$, see chapter 4 section 4.5.5. This interpretation is therefore presented in the discussion.

Table 5.3.4 Estimates of the seropositivity rate $\hat{p}^a(A)$ and the critical past-sensitivity $\hat{\eta}_c(A)$ for ages 9.7-9.8 yr (survey 1973) and for ages 9.8-10.4 yr (survey 1974). Central Jungle Area, Alalaparoe.

survey 1973			survey 1974		
age	seropos. rate	critical past-sens.	age	seropos. rate	critical past-sens.
years	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$	years	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$
9.7	0.56	0.79	9.8	0.41	0.87
9.8	0.57	0.64	9.9	0.42	0.70
			10.0	0.43	0.61
			10.1	0.43	0.55
			10.2	0.44	0.52
			10.3	0.45	0.49
			10.4	0.45	0.47

The spline-trend $\hat{\lambda}_s^a(t)$ is decreasing in period 9.7-9.8 yr before survey date - survey 1973 - and in period 9.8-10.4 yr before survey date - survey 1974 -.

Table 5.3.5 Estimates of the seropositivity rate $\hat{p}^a(A)$ and the critical past-sensitivity $\hat{n}_c^a(A)$ of children born in periods in which the spline-trend $\hat{\lambda}_s^a(t)$ is increasing ($p \leq 0.05$, one-sided test).
Central Jungle Area, Alalaparoe.

survey 1973			survey 1974		
age	seropos. rate	critical past-sens.	age	seropos. rate	critical past-sens.
years	$\hat{p}^a(A)$	$\hat{n}_c^a(A)$	years	$\hat{p}^a(A)$	$\hat{n}_c^a(A)$
2.8	0.29	0.67	2.7	0.23	0.23
3.0	0.32	0.57	2.8	0.23	0.23
3.5	0.40	0.48	2.9	0.23	0.23
4.0	0.40	0.43			
4.5	0.40	0.40			
14.8	0.91	0.98	21.5	0.86	0.96
15.0	0.92	0.98	22.0	0.86	0.96
17.0	0.93	0.97	23.0	0.88	0.96
19.0	0.94	0.97	24.0	0.89	0.96
21.0	0.95	0.97	25.0	0.90	0.96
23.0	0.96	0.96	26.0	0.92	0.96
25.0	0.96	0.97	26.7	0.93	0.97
26.4	0.97	0.97			

5.3.4 Discussion

The history of malaria transmission in the localities where the field study was carried out has been reported by van der Kaay (1975). The available data of that study are compared with the results of the analysis presented here.

The two villages where the field study was carried out are situated in a district which has been known for its virtual absence of malaria. In 1958 the Anti Malaria Campaign (AMC) initiated a country wide malaria eradication campaign. This only confirmed the absence of malaria transmission as no malaria cases have been reported from the district up to and including 1974, except for one imported P.falciparum case in 1965. No parasite positive cases nor people with a palpable spleen were found in the survey of 1973 and 1974.

Application of the two-step procedure to the data from the Coastal Area - which is an area of very low or no transmission - would cause artefacts. Normally, the seropositivity rate $\hat{p}^a(A)$ tends to increase with age, the 1973 data does not show such a tendency which is required for application of the model. Moreover many zeros are recorded for the seropositivity rates $\hat{p}^a(A)$ of the 1974 survey (see table 5.3.1); under these circumstances calculation of the p-value for the detection of a decrease in the spline-trend $\lambda_s^a(t)$ would lead to artificially low values. This is due to the method of estimation used to assess the variance of the rate of change in the spline-trend $\hat{\lambda}_s^a(t)$. The estimate of the variance is a weighted linear combination of terms (see chapter 2, formula (2.18))

$$\frac{\hat{p}^a(A_j)}{1 - \hat{p}^a(A_j) n_j} \frac{1}{n_j} \quad (j=1, \dots, k).$$

Therefore seropositivity rates $\hat{p}(A_j)$ which are zero will contribute to an underestimation of this variance. Furthermore the normal approximation of the distribution of the rate of change $\hat{\lambda}_s^a(t)$ in the spline-trend becomes doubtful in such a situation. Detection of change in the trend of the transmission intensity in areas where transmission is extremely low require

specially adapted models. These models will need larger samples than the samples collected in the Coastal Area.

Upper Surinam River

The survey was carried out in Dang and Kambaloea, two villages situated on the bank of the Upper Surinam River. In 1958 the malaria eradication program was started. The early years of the campaign had little impact on the transmission of malaria, the slide positivity rate remained high and showed great fluctuations. A gradual introduction of surveillance activities by the mid-sixties indicated further that little progress was made during the period 1965-1968. It was only in 1968, when the important decision was made to include in the annual residual spraying campaign dwellings near the planting ground, that a further progress was made. From 1966 medicated salt had also been used as an additional attack measure till the end of 1973. At this time it was withdrawn due to epidemic falciparum malaria spreading through the interior.

The falciparum malaria epidemic which spread through the interior between 1972 and 1974 is neither indicated by the constant infection rate model (table 5.3.2) nor by the time-dependent transmission model. Furthermore, application of the two-step procedure did not provide evidence for a downward trend of the transmission intensity in the period around 1969. The very small sample sizes in the youngest age groups and the width of the age-intervals may partly explain why the procedure did not yield significant results in these recent periods before survey date.

According to records on spleen rate and parasite rate collected in 1911 and 1939, the malaria endemicity before the start of the malaria eradication program may be classified as hyperendemic to holoendemic. The level of the

spline-trend $\hat{\lambda}_s^a(t)$, however, based on the data of the 1973 survey and 1974 survey, did not indicate a high degree of malaria endemicity before 1958. The main factor responsible for these biased results in the earlier period is probably the factor fading of antibodies. Furthermore it can not be excluded that the migration pattern of the adults in the population has influenced the results. The area is inhabited by the Saramaccaner tribe, the largest among the bush negroes. According to customary agricultural practice the women are cultivating the land. In this community the women and with her, the children have at least two homes. A more permanent one located in the village to which they belong by birth and a second, more temporary one, near the planting ground. The adult men spend most of their time along the coast in search of work, occasionally returning home. As a consequence of this social structure, it might be that the part of the curve $\hat{\lambda}_s^a(t)$ referring to the period before 1958 is also reflecting the malaria endemicity elsewhere.

Stoelmans Island

The study had been carried out along the Tapanahony river and the immediate surroundings of Stoelmans Island. What has been written about the inhabitants of Dang and Kambaloea is in its generality also applicable to the study area of Stoelmans Island. Hence there are doubts with respect to the applicability of the two-step procedure in this situation; the assumption that a homogeneous population is being dealt with may not be acceptable.

A statistically significant decrease in $\lambda_s^a(t)$ could not be established in the survey of 1973 as well in the survey of 1974. Only the 1973 survey provided some evidence for an increasing trend in $\lambda_s^a(t)$ in the years 1946, 1947, 1948 and at the end of 1971. These increases, however, could not be

substantiated further using the concept of critical past-sensitivity. Therefore a definite interpretation of the course of the spline trends is hardly possible. The shift in R-values of 1973 and 1974 - 8.4% \pm 1.3% in 1973 to 3.8% \pm 0.8% in 1974 (table 5.3.2) - suggests a strong decrease of transmission in the interval between the two surveys. This shift however seems to be influenced by the fading of antibodies which might have caused that a considerable number of marginally seropositives in the 1973 survey became seronegative in the 1974 survey.

Central Jungle Area

The field study was carried out in Alalaparoe a village in the South-West of Surinam. The village is mainly inhabited by Trio amerindians. Though these people have only recently started to settle, they still like to travel in smaller or larger groups and thus can be away from the village for periods of several months. Visits to relatives in other settlements either located in the South of Surinam or across the border into Brazil is not uncommon. The serological data of table 5.3.1 and thus the derived spline-trends do not reflect the transmission intensity in the village Alalaparoe, but rather the average malaria risk of the "community Alalaparoe". It remains doubtful, however, if this community can be considered as homogeneous.

- Decrease of transmission -

The 1973 survey indicates a downward trend of the transmission intensity in the year 1964.1-1966.6 (fig. 5.3.1) The two-step procedure applied to the 1974 data provides evidence for a decrease in transmission in the years 1965.0-1968.5 (fig. 5.3.2). Hence the methodology applied to the 1973 data and the 1974 data produces consistent results. The next question of course is as to which extent these results are in line with the history of malaria in

Alalaparoe.

Jan der Kaay (1975) reports that a health-post was opened in 1968, manned by a nurse to direct the medical and preventive care in the village. Table 5.3.6 presents the clinical malaria cases. Although the impression exists, that between 1965 and 1967 the inhabitants suffered much from malaria, no records are available for those years and unfortunately hard facts for the years 1965-1967 are not available. Application of the two-step procedure only confirms the impression of a relatively high degree of transmission in the years 1965-1967.

Period 9.7-9.8 yr before survey date (survey 1973) and period 9.8-10.4 yr before survey date (survey 1974) may be further considered for a decrease of transmission. If the actual past-sensitivity $\eta(A)$ of the children born in these periods would be smaller than the critical past-sensitivity $\hat{\eta}_c(A)$ presented in table 5.3.4, then these subperiods may be added to periods of decrease already established. It is very difficult to select ages for which $\eta(A) < \hat{\eta}_c(A)$. The past-sensitivity for age 9.8 years (1974 survey) might be smaller than 0.87. This would mean an additional decrease in the spline-trend of the transmission intensity in the last months of the year 1964. These considerations are of course not important from a practical point of view.

Table 5.3.6 Clinical malaria cases for the period 1968-1971 and laboratory confirmed cases for the period 1971-1974 in Alalaparoe.

year	1965	1966	1967	1968	1969	1979	1971	1972	1973	1974
population	320	325	333	390	419	438	468	472	445	468
no of malaria cases	?	?	?	33	58	18	0	313	2	0

Source: *van der Kaay, H.J. (1975)*. Malaria in Surinam, a sero-epidemiological study.

Adding or not adding such small periods to periods of decrease already found, would not change the picture of a downward trend of the transmission intensity indicated in the years 1964-1968.

- Increase of transmission -

If it could be assumed that the past-sensitivity $n(A)$ is not decreasing with age for children born in the periods where the spline trend $\lambda_s^a(t)$ is increasing (see table 5.3.3), then table 5.3.5 might be used to correct for the factor past-sensitivity. An increase in the spline trend $\lambda_s^a(t)$ is interpreted as a real increase if the actual past-sensitivity $n(A)$ happens to be larger than the estimates of the critical past-sensitivity $n_c(A)$ presented in table 5.3.5.

With respect to the 1973 survey the past-sensitivity $n(A)$ might be larger than the critical past-sensitivity for ages 3.0-4.5 yr, indicating an upward trend of the transmission intensity from the middle of 1969 to the end of 1970. The critical past-sensitivity for the ages 2.7-2.9 yr of the 1974 survey coincides with the seropositivity rate (see table 5.3.5). So the past-sensitivity $n(A)$ for these ages can be considered to be larger than $n_c(A)$. Consequently the 1974 survey provides some evidence for an increase in transmission intensity around 1972.

Periods of increase in $\hat{\lambda}_s^a(t)$ are also found in period 14.8-26.4 yr before survey date (1973 survey) and in period 21.5- 26.7 yr before survey date (1974 survey). The estimates of the critical past-sensitivity, however, appear to be very high. It seems difficult to decide if the actual past-sensitivity $n(A)$ is larger than $\hat{n}_c(A)$. For that reason it should be concluded that the course of the real transmission intensity remains uncertain in these periods.

From table 5.3.6 it seems indicated that malaria had not been a major

health problem in Alalaparoe until an epidemic outbreak in 1972. The epidemic was caused by *P.falciparum* and was rapidly brought under control as a result of a coordinated effort by the AMC and MZS (Stichting Medische Zending Suriname). The period of increase around 1972 indicated by the 1974 survey is in line with the epidemic in 1972. However the increasing trend of $\hat{\lambda}_s(t)$ based on the serological data of the 1973 survey is located two years before the actual increase of the transmission intensity. Taking into consideration the relatively large and unequal distances between the knots (the distances between the knots in period 0.0-7.5 yr before survey date are respectively 1.5 yr, 2 yr and 4 yr) it cannot be expected that the spline-trend $\lambda_s(t)$ can be used as a tool to determine exact annual trends. Therefore it might be that the indicated periods of increase of the spline-trends of both surveys may reflect the epidemic outbreak in 1972.

Final remarks

- a. Van der Kaay reports a very high specificity of the IHA test, provided that the discriminative titre level is taken as 1:40. The specificity of the IHA test in the Surinam study is indicated by the examination of 237 serum samples of healthy blood donors from Paramaribo between 18 and 69 years old; three individuals had a positive titre of 1:40 and one man of 65 years had a titre of 1:160. The possible relationship with a past history of malaria could not be further determined in these cases. The specificity of the test which is reflected in these findings indicate that it was acceptable to disregard nonspecific serological reactions in the epidemiological studies of the interior.
- b. The two-step procedure can not be applied if there are age groups with a seropositivity rate of 100%. The serological data from Alalaparoe did show 100% seropositives in the elder age groups (table 5.3.1). In order to

process the data in a standardized way one individual in each of the three age groups with 100% seropositives was converted to negative. This modification is a rather crude procedure, however, it is a very simple one acceptable in the analysis performed here. More preferable modifications which take into account the sample sizes of the age groups are discussed in Appendix 2.

- c. The ability of the two-step procedure for the detection of a downward trend in transmission intensity depends on the sample sizes in the age groups and the position of the knots of the spline approximation. Both factors are far from optimal in the Surinam study. Smaller distances between the knots and larger sample sizes are generally needed to obtain a more detailed picture of the trend in malaria transmission.

5.4 SEROLOGICAL DATA COLLECTED IN SRI LANKA

5.4.1 *Introduction*

The analysis of the serological data from Sri Lanka, already started in chapter 3, section 3.4, is continued in this section. The main objective is the detection of a downward trend of the transmission intensity while taking into account the factor of past-sensitivity. An attempt is made to combine the results of the serological surveys in order to assess the overall trend of the transmission intensity in Kilinochchi. The results of the application of two tentative methods of correcting for the factor past-sensitivity in a non-homogeneous population, introduced in chapter 4 section 4.5.4, are presented.

In order to evaluate the validity of the results a comparison is made with the results obtained from the retrospective parasitological study described in chapter 3, section 3.4.4.

5.4.2 Trend of the transmission intensity in the sub-areas

The two-step procedure for the detection of a downward trend in transmission intensity has been applied to the serological data from each sub-area (see chapter 3, table 3.2). The results are presented in 8 subfigures of fig. 5.4.1 (see page 186 and 187). The spline approximations $\hat{\lambda}_s^a(t)$ are shown in the upper-half of the subfigures. The distance between the knots of the spline approximation is one year. The sub-areas no. 1, 2, 3 and 8 show periods before survey date with a significant decrease in the spline approximation $\hat{\lambda}_s^a(t)$. These periods - result of step 1 of the procedure - are presented in table 5.4.1. This table also provides a further classification into subperiods according to diagram 4.4 (chapter 4). A decrease in the spline-trend of the real transmission intensity $\lambda_s(t)$ - step 2 of the procedure - is indicated in those subperiods in which the rate of change of the spline approximation $\hat{\lambda}_s^a(t)$ is sufficiently large ($\hat{\gamma}_s^a(t) > \hat{u}(t)$, see table 5.4.1). A downward trend of the transmission function is found in sub-area no. 1, 3 and 8 (fig. 5.4.1).

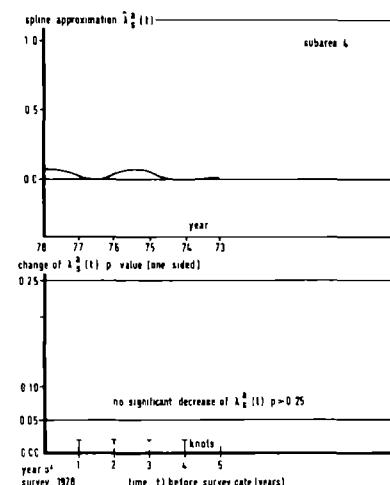
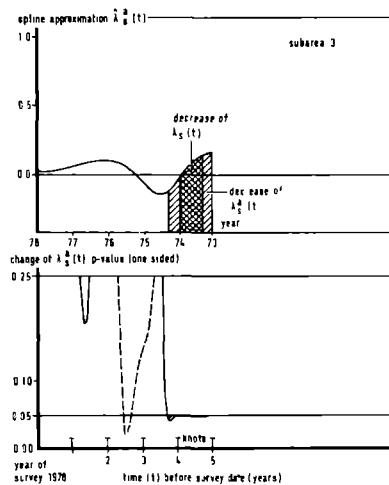
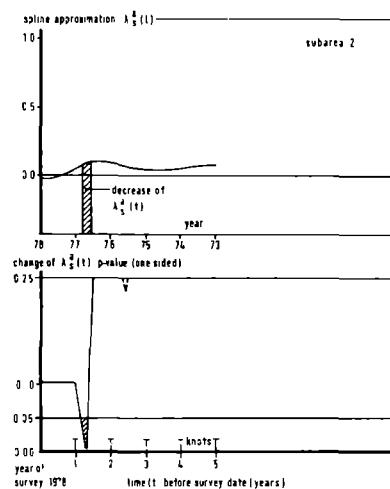
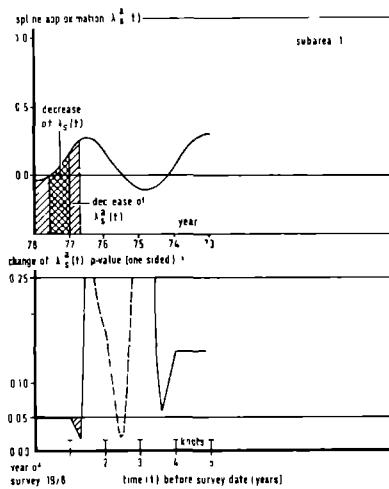
Table 5.4.1 Analysis by sub-area of serological data collected in the Kilinochchi district. Periods before survey date, with a significant decrease in spline approximation $\hat{\lambda}_s^a(t)$, classified into subperiods according to diagram 4.4.

Periods before survey date (yr)

Sub-area no	decrease of $\hat{\lambda}_s^a(t)$ $P \leq 0.05$	Classification into 4 subperiods		
		$\hat{\lambda}_s^a(t) < 0$	$\hat{\lambda}_s^a(t) \leq \hat{l}(t)$	$\hat{l}(t) < \hat{\gamma}_s^a(t) \leq \hat{u}(t)$
1	0.1-1.3	0.1-0.4		1.0-1.3 0.5-0.9
2	1.2-1.3			1.2-1.3
3	3.7-4.9	3.7-4.0		4.8-4.9 4.1-4.7
8	0.1-1.3	0.1-0.4		1.0-1.3 0.5-0.9

legenda: $\hat{\gamma}_s^a(t)$ =rate of change of $\hat{\lambda}_s^a(t)$, $\hat{l}(t)$ =lowerbound, $\hat{u}(t)$ =upperbound.

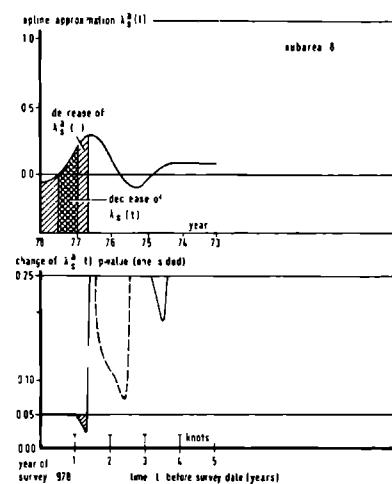
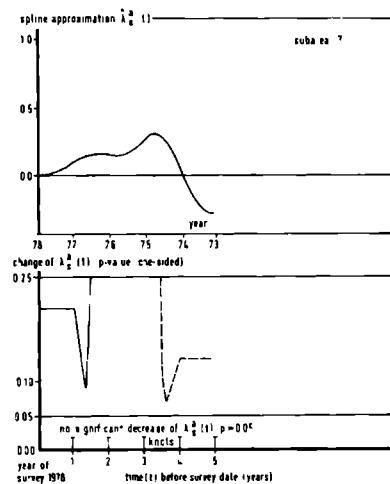
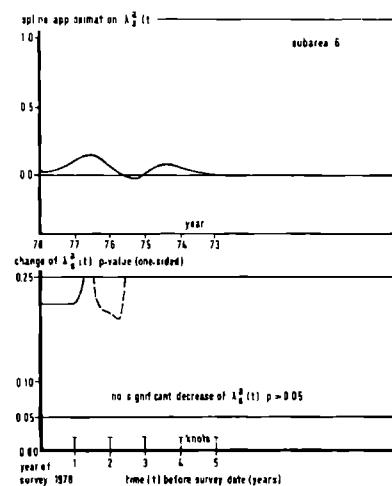
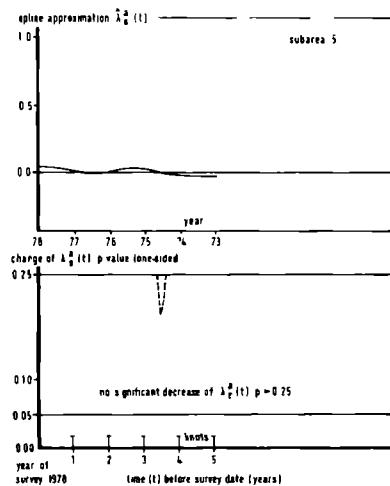
Figure 5.4.1 Analysis of serological data collected in Sri Lanka; detection of a downward trend of the transmission intensity in 8 sub-areas of the Kilinochchi district.



See general legenda page 247:

$\lambda_g^{\alpha}(t)$ = spline-trend of the antibody related transmission function

$\lambda_s(t)$ = spline-trend of the transmission intensity



Critical past-sensitivity

- a) Table 5.4.1 shows 4 subperiods in which the rate of decrease $\hat{\gamma}_s^a(t)$ of the spline approximation $\hat{\lambda}_s^a(t)$ takes on values between the lowerbound $\hat{\ell}(t)$ and the upperbound $\hat{u}(t)$. The estimates of the critical past-sensitivity for the corresponding age intervals are presented in table 5.4.2.
- b) The dashed one sided p-value curves take on values below 0.05 in sub-area no. 1 and sub-area no. 3 (see the subfigures of fig. 5.4.1). The periods before survey date in which $\hat{\lambda}_s^a(t)$ is increasing are 2.4-2.5 yr (sub-area no. 1) and 2.5-2.6 yr (sub-area no. 3). The estimates of the critical past-sensitivity $\hat{\eta}_c(A)$ and the seropositivity rates $\hat{p}^a(A)$ of children born in these periods are presented in table 5.4.3.

Table 5.4.2 Kilinochchi district; estimates of the seropositivity rate $\hat{p}^a(A)$ and the critical past-sensitivity $\hat{\eta}_c(A)$ in four sub-areas¹⁾

Sub-area no. 1			Sub-area no. 2		
age (yr)	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$	age (yr)	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$
1.0	0.03 ¹⁾	0.16	1.2	0.02	0.03
1.1	0.05	0.13	1.3	0.03	0.04
1.2	0.08	0.12			
1.3	0.10	0.12			

Sub-area no. 3			Sub-area no. 4		
age (yr)	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$	age (yr)	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$
4.8	0.13	0.23	1.0	0.03	0.06
4.9	0.14	0.15	1.1	0.05	0.09
			1.2	0.07	0.10
			1.3	0.09	0.10

¹⁾ The spline approximation $\hat{\lambda}_s^a(t)$ is decreasing, see table 5.4.1, in the corresponding periods before survey date.

Table 5.4.3 Kilinochchi district; estimates of the seropositivity rate $\hat{p}^a(A)$ and the critical past-sensitivity $\hat{r}_c(A)$ in two sub-areas¹⁾.

Sub-area no. 1			Sub-area no. 3		
age (yr)	$\hat{p}^a(A)$	$\hat{\eta}_c(A)$	age (yr)	$\hat{p}^a(A)$	$\hat{r}_c(A)$
2.4	0.25 ¹⁾	0.25	2.5	0.13	0.15
2.5	0.25	0.25	2.6	0.14	0.15

1) The spline approximation $\hat{\lambda}_s^a(t)$ is increasing in the corresponding periods before survey date.

The interpretation of the values of the critical past-sensitivity $\hat{r}_c(A)$ depends on the subjective comparison of $\eta(A)$ with $\hat{\eta}_c(A)$, see chapter 4 section 4.5.5. This interpretation is therefore presented in the discussion (section 5.4.4).

5.4.3 Assessment of the overall trend

In order to assess the overall trend of the transmission intensity in Kilinochchi, the results of the serological surveys in the sub-areas have to be combined. Table 5.4.4 (see page 190) presents the crude proportions age related seropositives of the total sample. The two-step procedure for a homogeneous population has been applied to the data of this table. Fig. 5.4.2 (see page 191) presents the results.

A decrease of the spline approximation $\hat{\lambda}_s^a(t)$ is found in period 0.0-1.4 yr before survey date ($p \leq 0.05$, one-sided test). This period is further analysed in step 2 of the procedure. A downward trend of the transmission intensity is indicated in period 0.2-1.1 yr before survey date (decrease of $\lambda_s(t)$).

Table 5.4.4 crude proportions of seropositives in all serum samples collected in the Kilinochchi district (IFA test, P.field1, positive is titre $\geq 1:80$).

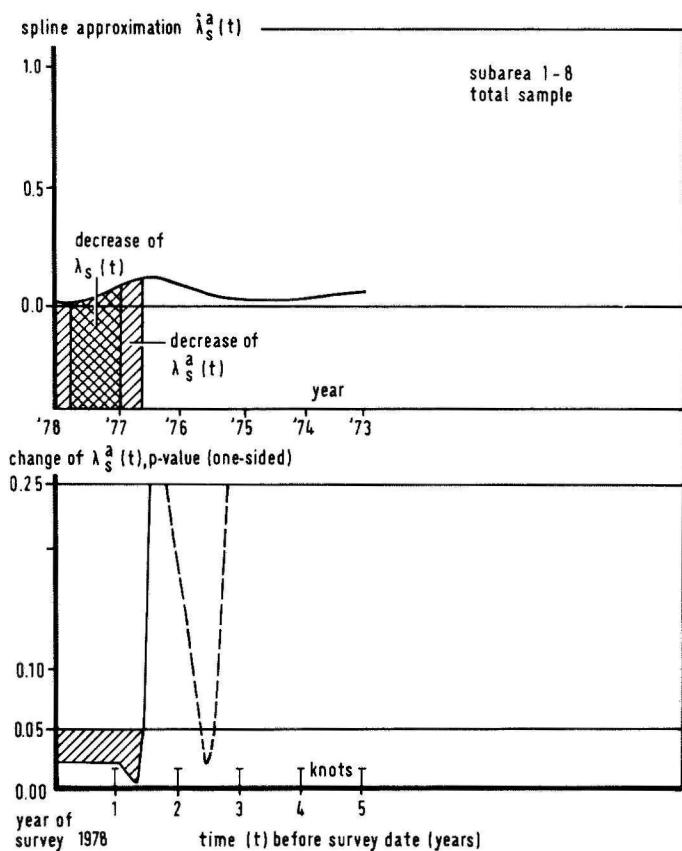
age group (yr)	no exam.	no pos.	% pos.
0.5-1.5	267	9	3.4
1.5-2.5	350	47	13.4
2.5-3.5	370	66	17.8
3.5-4.5	342	68	19.9
4.5-5.5	349	84	24.1

An increase of the spline approximation $\lambda_s^a(t)$ of the antibody related transmission function is found in period 2.3-2.5 yr before survey date (fig. 5.4.2, $p \leq 0.05$, one-sided test). The estimates of the seropositivity rate $p^a(A)$ of children born in this period - i.e. 2.3 yr, 2.4 yr, 2.5 yr - are respectively 0.15, 0.15, 0.16. The estimates of the critical past-sensitivity for these ages are: 0.19, 0.18 and 0.18 respectively.

Two tentative methods were proposed in chapter 4, section 4.5.4, for the detection of a downward trend of the transmission intensity in a non-homogeneous population. These methods are applied here. Both methods are based on the application of the spatial and time-dependent transmission model followed by a correction procedure for the factor past-sensitivity. Step 1 is the same for both methods. In chapter 3, section 3.4.3, an overall downward trend in $\lambda_s^a(t)$ was indicated in period 0.0-1.4 yr before survey date ($\lambda_s^a(t)$ is decreasing, $p \leq 0.05$, one-sided test). This period before survey date is now the object of further analysis.

The first tentative approach uses a formal extension of the maximal linear correction (MLC) method. The rate of change $\hat{\Gamma}_s^a(t)$ of the overall trend $\hat{\Lambda}_s^a(t)$ happens to be larger than the formal upperbound $\hat{U}_s(t)$ in period

Figure 5.4.2 Analysis of serological data collected in Sri Lanka; assessment of the overall trend of the transmission intensity in the Kilinochchi district using the crude proportions seropositives.



See general legenda page 247:

$\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

$\lambda_s(t)$ = spline-trend of the transmission intensity

0.2-1.1 yr before survey date, indicating an overall downward trend of the transmission intensity in this period.

The second tentative method is based in the application of the MLC method for the factor past-sensitivity in each sub-area. Table 5.4.5 presents, for each time point t in period 0.0-1.4 yr before survey date, those sub-areas which show a decreasing spline approximation $\hat{\lambda}_s^a(t)$ and fulfil the condition $\hat{\gamma}_s^a(t) > \hat{u}(t)$. A downward trend of the transmission intensity is indicated in period 0.5-0.9 yr before survey date in the sub-areas no. 1, 3, 6, 7 and 8.

5.4.4 Discussion

It will be argued that the analysis on sub-area level as well as the overall analysis has produced valid results in this application. In view of DDT resistance to the local vector *A.culicifacies*, DDT was replaced by malathion in September-October 1977 i.e. one year before the date of the serological survey. Therefore establishing a downward trend of the transmission intensity in a period of about one year before survey date, while taking into account the factor of past-sensitivity, would provide evidence for the effectiveness of the substitute insecticide in the area Kilinochchi.

a) Analysis by sub-area

The patchy nature of malaria transmission in Kilinochchi dominates the evaluation of any malario metric figure. The 8 sub-areas of Kilinochchi do not show the same serological profile (see chapter 3, table 3.1); therefore an analysis by sub-areas seems indicated in the first instance. Application of the two-step procedure to the serological data from each sub-area indicates a downward trend of the transmission intensity in period 0.5-0.9 yr before

Table 5.4.5 Analysis of serological data collected in 8 sub-areas of the Kilinochchi district. Assessment of the overall trend in transmission intensity; maximal linear correction (MLC) for the factor of past-sensitivity by sub-area.

time before survey date (yr)	number of sub-areas with decrease $\lambda_s^a(t)$	$\hat{\lambda}_s^a(t)$	sub-area no. with indication of real decrease (x)	1	2	3	4	5	6	7	8
p ≤ 0.05											
0.1	6								x		
0.2	6								x		
0.3	6			x				x	x		
0.4	6			x			x	x	x		
<hr/>											
0.5	6		x	x			x	x	x		
0.6	6		x	x			x	x	x		
0.7	6		x	x			x	x	x		
0.8	6		x	x			x	x	x		
0.9	6		x	x			x	x	x		
<hr/>											
1.0	6			x			x	x			
1.1	6			x					x		
1.2	6				x						
1.3	6					x					
1.4	5										

MLC correction: rate of decrease $\hat{\lambda}_s^a(t) > \hat{u}(t)$.

survey date in sub-area no. 1 and sub-area no. 8 (fig. 5.4.1, decrease of $\lambda_s^a(t)$). The moving annual slide positivity rate (SPR) obtained from the retrospective parasitological study in the sub-areas no. 1 and 8 confirms this decreasing tendency (see chapter 3, fig. 3.6 and table 3.7). The indicated decrease in $\lambda_s^a(t)$ in sub-area no. 3 in period 4.1-4.7 yr before survey date, however, is not in line with the SPR trend in this sub-area.

The change of $\hat{\lambda}_s^a(t)$ in period 2-5 yr before survey date may be an artefact, to a large extent induced by the relatively low seropositivity rate (5%) in the age group 3.5-4.5 yr in sub-area no. 3 (see chapter 3, table 3.1).

Critical past-sensitivity

In four sub-areas (including sub-area no. 3) periods before survey date are found in which $\hat{\lambda}_s^a(t)$ is decreasing and in which the rate of decrease $\hat{\gamma}_s^a(t)$ of the spline approximation $\hat{\lambda}_s^a(t)$ takes on values between the lower-bound $\hat{l}(t)$ and the upperbound $\hat{u}(t)$ (see table 5.4.1). It is not likely that the past-sensitivity $n(A)$ of the serological test for children born in these periods assumes a value less than the estimate of the critical past-sensitivity $n_c(A)$ presented in table 5.4.2. Hence a definite interpretation of the course of $\hat{\lambda}_s^a(t)$ in these periods seems not possible and the course of $\lambda_s^a(t)$ remains uncertain in these periods (cf. chapter 4, diagram 4.4).

With respect to the detection of an upward trend of the transmission intensity in each sub-area, an increase in $\hat{\lambda}_s^a(t)$ is found in sub-area no. 1 and sub-area no. 3 (fig. 5.4.1). For reasons already mentioned the increase in sub-area no. 3 in period 2.5-2.6 yr before survey date should be disregarded. The increase in $\hat{\lambda}_s^a(t)$ in sub-area no. 1 2.4-2.5 yr before survey date might be the result of a decreasing past-sensitivity $n(A)$ for children born in this period. On the other hand if $n(A)$ is not decreasing with age the increase in $\hat{\lambda}_s^a(t)$ may reflect a real increase in the trend of the transmission intensity. The past-sensitivity $n(A)$ of the serological test for children born in this period is surely larger than the estimates of the critical past-sensitivity presented in table 5.4.3, thereby providing some evidence for a real increase in $\lambda_s^a(t)$ (cf. chapter 4, diagram 4.5). The course of the SPR trend in sub-area no. 1 shows an upward trend about 2.75 yr before survey

date (chapter 3, fig. 3.6 and table 3.7), indicating that this increase in $\hat{\lambda}_s^a(t)$ may reflect reality.

b) *Overall analysis*

A disadvantage of only an analysis by sub-area is the relative limited ability to detect a downward trend of the transmission intensity in a particular sub-area. Nearly all spline approximations $\hat{\lambda}_s^a(t)$ in the sub-areas may show the same tendency in a certain period before survey date while a significant change of $\hat{\lambda}_s^a(t)$ in a particular sub-area is not indicated. In situations like this an overall analysis should be performed additionally to the sub-area analysis, pooling the evidence obtained from the sub-areas. Three methods of combinations have been attempted: a crude method based on the proportions seropositives in the total sample and the two other tentative methods described in chapter 4, section 4.5.4. Each method will generally lead to a different result as each is based on a different function of the observations. The common objective, however, is the detection of a downward trend of the transmission intensity while taking into account, to some extent, the factor past-sensitivity.

Application of the two-step procedure to the crude proportions seropositives of the total sample may produce artefacts in case we are dealing with a non-homogeneous population. Even an analysis based on unbiased estimates of the age related cumulative inoculation rates $p(A)$ might be misleading (see chapter 3, section 3.3.2). In this application it appears that the crude method produces exactly the same results as the first tentative method of correcting for the factor past-sensitivity in a non-homogeneous population. This is due to the fact that the crude seropositivity rate is approximately the same as the average seropositivity rate of the eight

sub-areas (see chapter 3, section 3.5.1). This implies that the spline approximation $\hat{\lambda}_s^a(t)$ based on the crude seropositivity rate is approximately identical to the average antibody related transmission function $\hat{\Lambda}_s^a(t)$. Hence the crude method produces practically the same results as application of the spatial and time-dependent transmission model with maximal linear correction (MLC) for the factor of past-sensitivity. The results of these methods - a downward trend of the transmission in a period of about a year immediately before survey date - are in line with the parasitological trend based on the quarterly slide positivity rates in this period (see chapter 3, fig. 3.7 and table 3.7).

The second tentative method can be considered as a combination of an overall and local analysis. The results of the first step (overall analysis) is followed by maximal linear correction on sub-area level (local analysis). This type of analysis will generally lead to a larger number of sub-areas - as compared with a local analysis - in which a decrease in the trend of the transmission is indicated, provided that there are no strong interactions between the spline approximations $\hat{\lambda}_s^a(t)$ in the sub-areas. In this particular application the overall analysis adds the sub-areas no. 3, 6 and 7 to the sub-areas no. 1 and no. 8 already found in the analysis by sub-area (table 5.4.5). The indicated downward trend of the transmission intensity in the sub-areas no. 3 and no. 6 is confirmed by the malario metric data (see chapter 3, fig. 3.6 and table 3.7). Sub-area no. 3 has not been excluded here because the course of $\hat{\lambda}_s^a(t)$ immediately before survey date is not strongly influenced by the low seropositivity rate in the age group 3.5-4.5 yr.

- Detection of an upward trend of the transmission intensity -

Generally periods of increase in $\hat{\lambda}_s^a(t)$ are difficult to interpret. In this application, however, the increase in $\hat{\lambda}_s^a(t)$ in the middle of 1976

seems to reflect a real upward trend of the transmission intensity. The overall trend of the slide positivity rate two years before survey date (chapter 3, fig. 3.7 and table 3.7) supports this interpretation.

5.5 NIGERIA, GARKI-PROJECT

5.5.1 *Introduction*

The *Garki research project* on the epidemiology and control of malaria in the northern part of Nigeria, conducted jointly by the World Health Organization and the government of Nigeria, is probably the most extensive research project on malaria ever performed. As part of this project a longitudinal sero immunological investigation, including 8 serological surveys, was carried out from 1971 to 1975. The project included a preintervention phase, an intervention phase with intensive malaria control and a post intervention phase. The serological study covered one control village cluster and two village clusters receiving the most intensive treatment.

Dr. L. Molineaux - Epidemiological Methodology and Evaluation Malaria Action Programma WHO - has made available serological data of survey no. 2 and survey no. 8, for an analysis with the methods developed in the preceding chapters. The main objectives of this preliminary study are to investigate whether the two-step procedure, described in chapter 4 section 4.2.4, applied to the serological results of the IHA test, is able to determine a downward trend of the transmission intensity in the intervention period in the villages where malaria control was carried out; and to verify whether application of the methodology to the serological data of the unprotected population does not indicate such a trend during this period.

5.5.2 Serological data and study design

A brief summary, based on the article of Brögger *et al.* (1978), is given regarding the setting of immunological survey no. 2 and immunological survey no. 8 in the study design of the Garki project. The project involved a 5-year longitudinal study of eight clusters of 2 - 4 villages each. The study comprised three phases: 1½ years of baseline observations (dry and wet season of 1971 and dry season of 1972), 1½ years of intervention (wet season of 1972, dry and wet season of 1973) and 2 years of follow-up. During the intervention phase, two village clusters were kept as controls and two are assigned to each of three malaria control methods. The serological study covered one control village cluster and the two clusters receiving the most intensive control (spraying of residual insecticide and mass drug administration). The two village clusters with malaria control were each composed of two villages and a section of a third, and their registered populations at the first survey were 982 and 828; the control cluster was composed of two villages with a population of 1,148.

Comprehensive entomological and parasitological observations were made. Parasitological surveys, aiming at total coverage were conducted every 10 weeks, except in the dry seasons of 1974 and 1975; 23 parasitological surveys were carried out. The 8 serological surveys coincided with selected parasitological surveys and also aimed at total coverage. Six immunological parameters were studied (IgG, IgM, Ouchterlony test, IFA, IHA and ELISA).

Immunological survey no. 2 was performed in May 1972 just before the intervention period (dry season, base line period). The results of the IHA test with *P.falciparum* as antigen are presented in table 5.5.1 (up to 43 yr). In order to investigate the influence of the choice of the discriminative titre level of the IHA test on the results of the analysis two definitions of "serological positive" have been used, i.e. titre $\geq 1:16$ and titre $\geq 1:32$.

Table 5.5.1 Serological data collected before intervention in the Garki district (Nigeria); immunological survey no. 2, IHA test (*P. falciparum* antigen), May 1972.

age group (yr)	no. exam.	<i>IHA test</i>		<i>pos. is titre $\geq 1:16$</i>		<i>pos. is titre $\geq 1:32$</i>	
		no. pos.	% pos.	no. pos.	% pos.		
< 1	34	23	68	19	56		
1 - 4	209	190	91	182	87		
5 - 8	309	290	94	280	91		
9 - 18	369	355	96	345	94		
19 - 28	338	332	98	329	97		
29 - 43	628	618	98	614	98		

Immunological survey no. 8 was carried out in the wet season of 1975, in the protected as well as in the unprotected populations about 21 months after the end of the intervention. The results of the IHA test (*P.falciparum* antigen) are shown in table 5.5.2 (see page 200).

5.5.3. Detection of a downward trend in the transmission intensity

The two-step procedure applied to the data of immunological survey no. 2 (IHA test), data collected before the intervention period in May 1972, does not provide evidence for a downward trend of the transmission intensity.

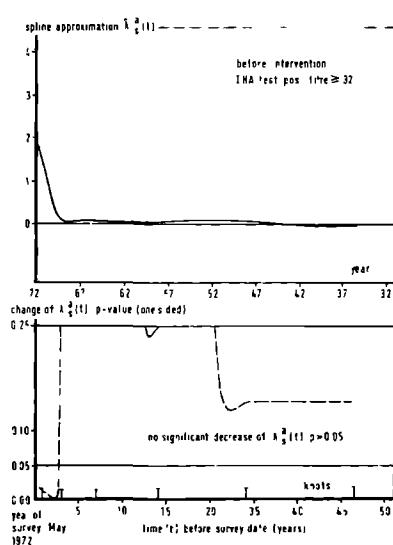
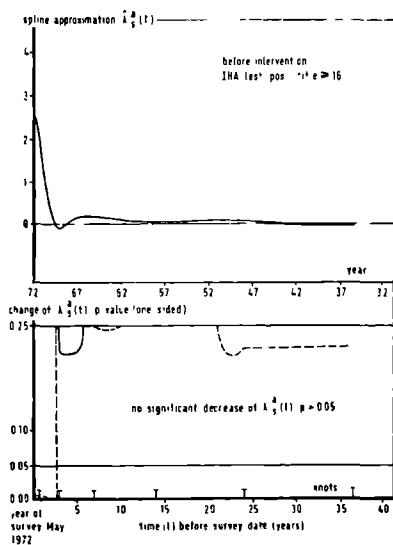
Fig. 5.5.1 (see page 201) shows in two sub-figures the spline approximations $\hat{\lambda}_s^a(t)$ based on the age related seropositive rates presented in table 5.5.1. Two definitions of "serologically positive" have been used; in both cases the one-sided p-value function for the detection of a decrease in the spline-trend $\lambda_s^a(t)$ - lower half of the figure - does not take on values below the critical level 0.05.

Table 5.5.2 Serological data of the previously protected and unprotected populations in the Garki district (Nigeria); immunological survey no. 8, IHA test (*P.falciparum* antigen), September/October 1975.

IHA test		pos. is titre $\geq 1:16$		pos. is titre $\geq 1:32$	
age group (yr)	no. exam.	no. pos.	% pos.	no. pos.	% pos.
<i>previously unprotected population</i>					
< 1	24	12	50	9	38
1 - 4	89	79	89	74	83
5 - 8	83	76	92	72	87
9 - 18	180	173	96	163	91
19 - 28	100	99	99	94	94
29 - 43	216	213	99	209	97
<i>previously protected population</i>					
< 1	50	42	84	34	68
1 - 4	219	194	89	177	81
5 - 8	169	161	95	155	92
9 - 18	299	292	98	286	96
19 - 28	152	151	99	150	99
29 - 43	420	416	99	412	98

The spline approximations $\hat{\lambda}_s^a(t)$ and corresponding p-value functions based on the proportions seropositives in the previously unprotected population and previously protected population in September/October 1975 are presented in subfigures of fig. 5.5.2 (see page 202). A decrease in $\hat{\lambda}_s^a(t)$ in the previously unprotected population is not found (one-sided test, $p > 0.05$). The previously protected population, however, shows with respect to both definitions of serologically positive a statistically significant decrease in $\hat{\lambda}_s^a(t)$. A decrease in the spline-trend $\lambda_s^a(t)$, i.e. step I of the procedure, is indicated in period 2.7-5.9 yr before survey date (positive is titre $\geq 1:16$)

Figure 5.5.7 Spline approximations $\hat{\lambda}_s^a(t)$ based on the age related seropositivity rates before intervention in May 1972 (immunologic survey no.2; IHA test, two definitions of serologically positive).

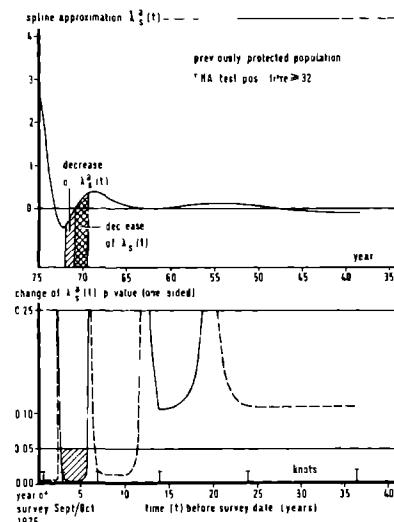
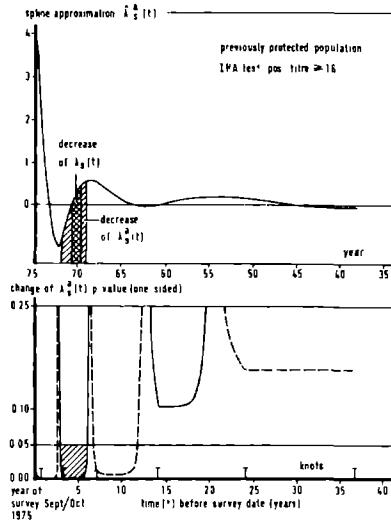
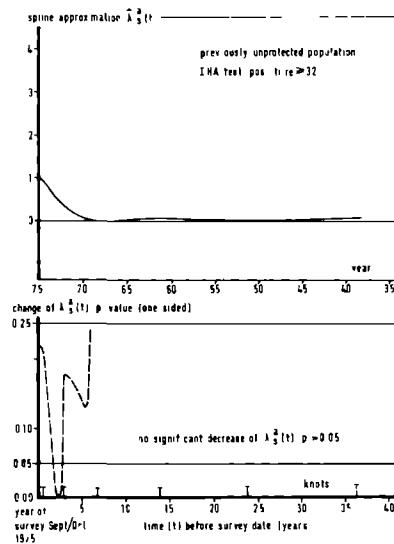
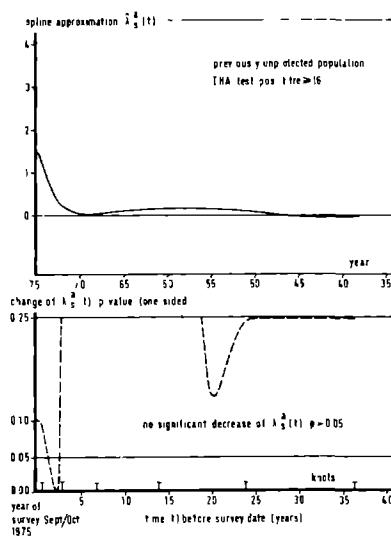


See general legenda page 247:

$\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

respectively 2.8-5.7 yr before survey date (positive is titre $\geq 1:32$). These periods are further analysed in step 2 of the procedure. The classification into subperiods according to diagram 4.4 (chapter 4) is presented in table 5.5.3 (see page 203). A downward trend in the transmission intensity is indicated in period 4.2-5.3 yr before survey date (positive is titre $\geq 1:16$) and 3.9-5.2 yr before survey date (positive is titre $\geq 1:32$).

Figure 5.5.2 Spline approximations $\hat{\lambda}_s^a(t)$ based on the age related seropositivity rates in the previously protected and unprotected populations (immunologic survey no. 8; IHA test, Sept/Oct 1975).



See general legenda page 247:

$\lambda_s^a(t)$ = spline-trend of the antibody related transmission function
 $\lambda_s(t)$ = spline-trend of the transmission intensity

Table 5.5.3 Analysis of serological data collected in the previously protected population in the Garki district (Nigeria); immunological survey no. 8, IHA test (*P.falciparum* antigen). Periods before survey date with a significant decrease in spline trend $\hat{\lambda}_s^a(t)$, classified into subperiods according to diagram 4.4.

Periods before survey date (yr)		
THA test	pos. is titre $\geq 1:16$	pos. is titre $\geq 1:32$
decrease $\hat{\lambda}_s^a(t)$, $p \leq 0.05$	2.7 - 5.9	2.8 - 5.7
Classification into subperiods		
$\hat{\lambda}_s^a(t) < 0$	2.7 - 4.1	2.8 - 3.8
$\hat{\gamma}_s^a(t) \leq \hat{\lambda}(t)$	5.4 - 5.9	5.4 - 5.7
$\hat{\lambda}(t) < \hat{\gamma}_s^a(t) \leq \hat{u}(t)$		5.3
$\hat{\gamma}_s^a(t) > \hat{u}(t)$	4.2 - 5.3	3.9 - 5.2

legenda: $\hat{\gamma}_s^a(t)$ =rate of change of $\hat{\lambda}_s^a(t)$, $\hat{\lambda}(t)$ =lowerbound, $\hat{u}(t)$ =upperbound.

5.5.4 Discussion

The two-step procedure indicated a downward trend of the transmission intensity from the middle of 1970 to the end of 1971 in the previously protected population whereas in the unprotected population a decrease in transmission could not be established. This result is roughly in line with the parasitological findings; a sharp decrease in the crude proportions positive for *P.falciparum* and *P.malariae* was observed in the wet season of 1972 in the protected population and such a decrease was not found in the unprotected population (Brögger et al. (1978)).

The mid-point of the period of decrease indicated by the two-step procedure is, however, situated more than one year before the actual decrease in the

transmission intensity which beyond any doubt occurred in the middle of 1972. This discrepancy between the parasitological findings and the result presented here may be explained, at least to some extent, by the fact that the analysis is based on a crude classification of the individuals into age groups. The spline approximation $\hat{\lambda}_s^a(t)$ describes the trend of the antibody related transmission function between the knots and these correspond with the mid-points of the age groups. So a detailed picture describing the change in the annual trend in the years 1971- 1975 cannot be expected on account of an analysis based on the age groups < 1 yr, 1-4 yr, 5-8 yr etc. A reanalysis based on the sero-positivity rates in smaller age groups - performed in a way as has been done with the Sri Lanka data - is needed in order to obtain a more refined picture. The final judgement on the applicability of the two-step procedure for the evaluation of malaria control measures in Garki-like situations should be postponed until the results of such a reanalysis are available.

It might be preferable not to put aside those time points in which the spline approximation $\hat{\lambda}_s^a(t)$ appears as negative. In step 1 of the procedure a decrease in the spline-trend $\hat{\lambda}_s^a(t)$ is found in the years 1970, 1971, 1972, 1973. The spline-trend $\hat{\lambda}_s^a(t)$ happens to be negative in the year 1972 and this is the year when the crude parasite rate showed a sharp decrease.

The choice of the discriminative titre level of the IHA test (titre \geq 1:16 or titre \geq 1:32) has had little influence on the results of this preliminary analysis. It would be very interesting to know whether an analysis based on the seropositivity rates in yearly age groups of children less 6 years old shows the same robustness. It would also be interesting to investigate whether the results of the other serological tests, especially the more sensitive IFA test, do provide the same picture with respect to a downward trend in transmission intensity.

After the intervention period the prevalence of *P.falciparum* remained

at low level in the dry season of 1974; in the wet season it rose rapidly to the control or baseline level. The resurgence of *P.malariae* was slower, and by the last parasitological survey in 1975 its prevalence was still clearly below the control level in the unprotected population (*Brögger et al. (1978)*).

The periods of increase of $\hat{\lambda}_s^a(t)$ immediately before survey date in the unprotected populations (fig. 5.5.1, fig. 5.5.2) do not reflect an upward trend of the transmission intensity; these periods of increase are clearly artefacts that might have been induced by the fading of maternal antibodies in newborns and/or a decreasing past-sensitivity $\eta(A)$ in the younger age groups. Comparison of the rate of increase of $\hat{\lambda}_s^a(t)$ in the protected population with the rate of increase of $\hat{\lambda}_s^a(t)$ in the unprotected population about one year before the survey date of immunological survey no. 8 (see fig.

5.5.2) suggest a difference in the rate of change of $\lambda_s^a(t)$; this could be indicative for an increase of transmission in the years 1974 and 1975 in the protected population as compared with the unprotected control population. However, beyond doubt the rate of increase of $\hat{\lambda}_s^a(t)$ in 1974 and 1975 in the previously protected population (fig. 5.5.2) is also to a very large extent an artefact due to the same reasons that apply to the unprotected populations. These results confirm the conception that increases in the spline approximation $\hat{\lambda}_s^a(t)$ are very difficult to interpret, especially when these periods are near to the survey date.

5.6 SEROLOGICAL DATA FROM PANAMA

5.6.1 Introduction

Serological data from Chepigana and Pinogana, two adjacent areas in Panama, have been placed at our disposal by Dr. Nájera - Chief Malaria, Parasitic Diseases and Vector Control, Pan American Health Organization -

so that these data could be analysed with the methods developed in the preceding chapters. One objective of this study was to verify whether application of the time-dependent transmission model produced the same result as those reported by Dr. Nájera. Further objectives were to apply the two-step procedure for the detection of a downward trend of the transmission intensity in each area and to combine the serological results of the two areas in order to assess the overall trend of the transmission intensity.

5.6.2 The trend of the transmission intensity in Chepigana and Pinogana

In Chepigana and Pinogana the IFA test has been applied in order to determine in both populations the seroreactivity to *P.vivax* and *P.falciparum* antigen. The serological surveys were carried out in 1977-1978 and the age related seropositivity rates in the two samples are presented in table 5.6.1 (*P.vivax* antigen) and table 5.6.2 (*P.falciparum* antigen). A titre $\geq 1:16$ was considered as positive.

Table 5.6.1 Results of the IFA test (*P.vivax* antigen) carried out on serum samples collected in Chepigana and Pinogana, positive is titre $\geq 1:16$.

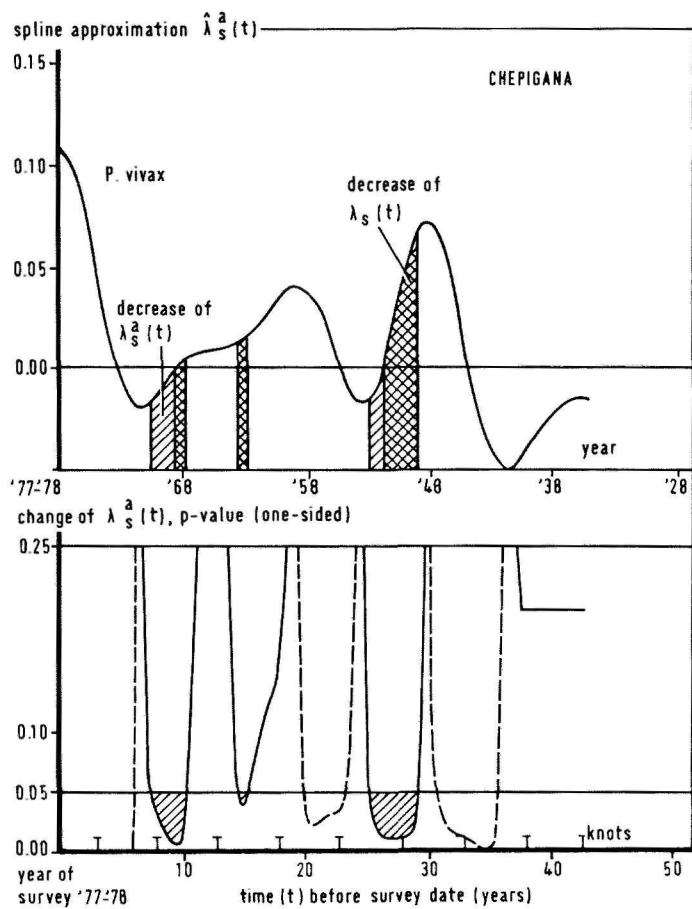
age group (yr)	Chepigana			Pinogana		
	no exam.	no pos.	% pos.	no exam.	no pos.	% pos.
1 - 4	641	147	23	264	69	26
5 - 9	795	179	23	370	111	30
10 - 14	546	124	23	252	75	30
15 - 19	355	102	29	197	62	32
20 - 24	237	92	39	185	74	40
25 - 29	225	84	37	123	52	42
30 - 34	171	90	53	87	39	45
35 - 39	171	76	44	88	33	38
40 - 44	129	48	37	71	28	39

Table 5.6.2 Results of the IFA test (*P.falciparum* antigen) carried out on serum samples collected in Chepigana and Pinogana, positive is titre $\geq 1:16$.

age group (yr)	Chepigana			Pinogana		
	no exam.	no pos.	% pos.	no exam.	no pos.	% pos.
1 - 4	641	68	11	264	25	10
5 - 9	795	120	15	370	39	11
10 - 14	546	101	19	252	19	8
15 - 19	355	89	25	197	42	21
20 - 24	237	71	30	185	42	23
25 - 29	225	81	36	123	43	35
30 - 34	171	56	33	87	32	37
35 - 39	171	85	50	88	37	42
40 - 44	129	67	52	71	37	52

The two-step procedure for the detection of a downward trend of the transmission intensity was applied to these data. The spline approximations $\hat{\lambda}_s^a(t)$ of the antibody related transmission functions, based on the seroreactivity to *P.vivax* antigen in Chepigana and Pinogana, are presented in fig. 5.6.1 (see pages 208 and 209). The spline approximations $\hat{\lambda}_s^a(t)$ based on the seroreactivity to *P.falciparum* antigen in the two populations are presented in fig. 5.6.2 (see pages 210 and 211). The knots of the spline approximations are situated 3.0, 7.5, 12.5, 17.5, 22.5, 27.5, 32.5 and 37.5 yr before survey date. The one-sided p-value functions for the detection of periods of decrease of $\hat{\lambda}_s^a(t)$ - step 1 of the procedure - are shown in the lower-half of the figures. The periods in which a downward trend of the transmission intensity is indicated - decrease of $\lambda_s(t)$, step 2 of the procedure - are presented in the upper-half of these figures.

Figure 5.6.1 Detection of a downward trend of the transmission intensity in Chepigana and Pinogana based on the seroreactivity to P.vivax antigen (IFA test).



See general legend page 241:

$\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

$\lambda_s(t)$ = spline-trend of the transmission intensity

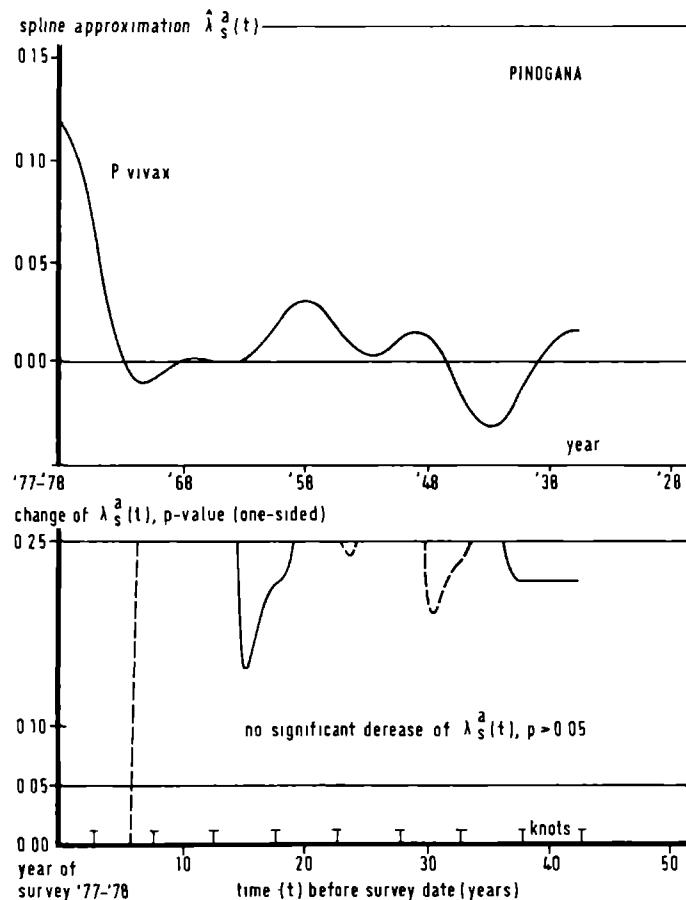
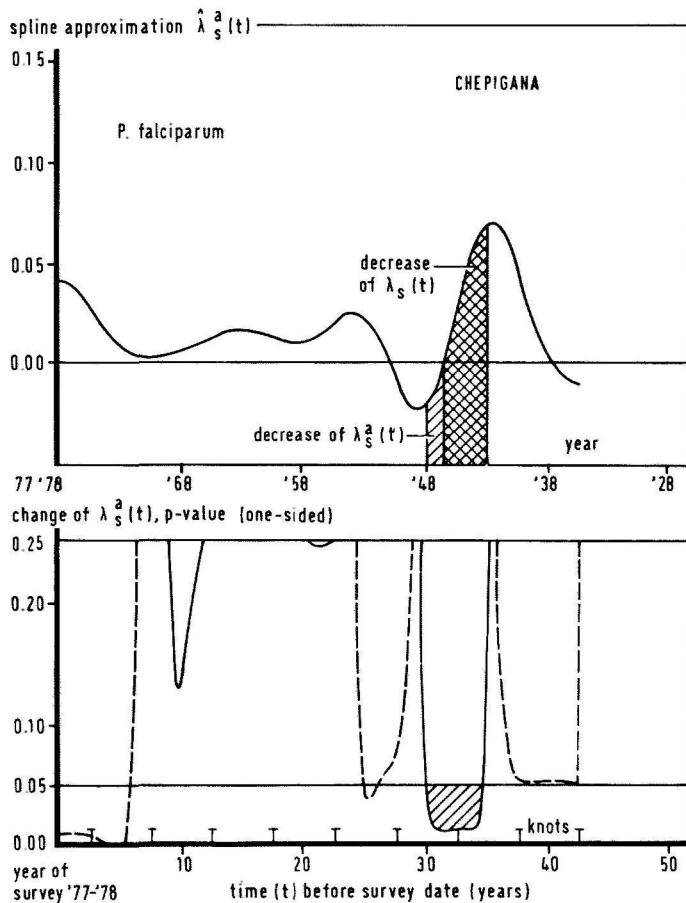


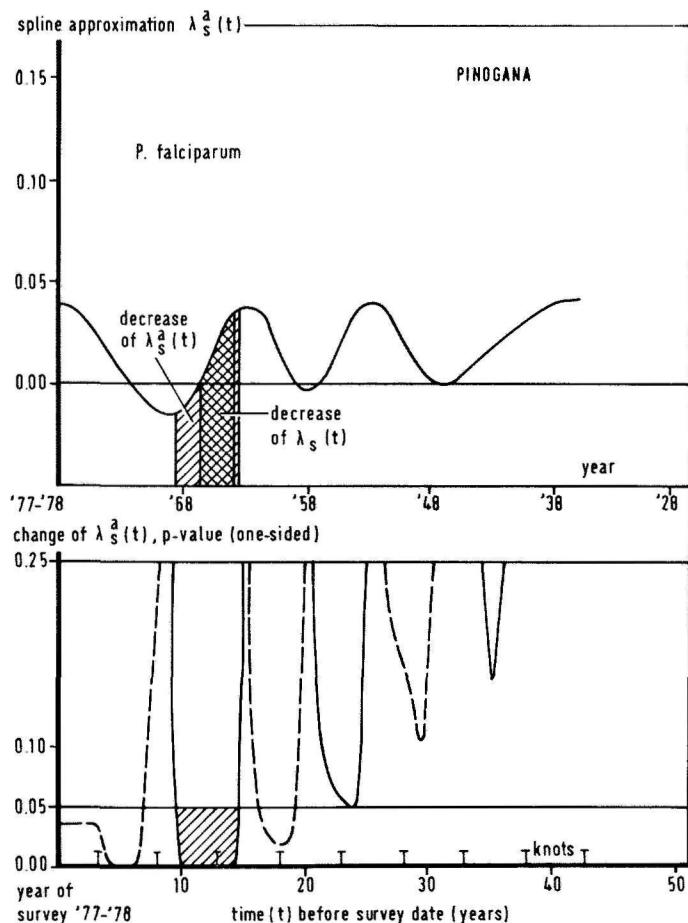
Figure 5.6.2 Detection of a downward trend of the transmission intensity in Chepigana and Pinogana based on the seroreactivity to *P.falciparum* antigen (IFA test).



See general legenda page 247:

$\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

$\lambda_s(t)$ = spline-trend of the transmission intensity



5.6.3 Assessment of the overall trend

The serological profiles of Chepigana and Pinogana presented in table 5.6.1 and table 5.6.2 are different. Comparison of the seropositivity rates by the use of the overall Chi-square test described in chapter 3, section 3.2, indicates a clear difference in seroreactivity (*P.vivax* antigen; $\chi^2 = 15.26$, $df = 9$, $p = 0.08$; *P.falciparum* antigen $\chi^2 = 24.16$, $df = 9$, $p = 0.04$). Further comparison of the seropositivity rates in Chepigana and Pinogana for each age group - Chi-square test for the comparison of two proportions - shows differences with respect to the seroreactivity in the age groups 5-9 yr resp. 10-14 yr (*P.vivax* antigen $p = 0.01$ resp. $p = 0.04$; *P.falciparum* antigen $p = 0.04$ resp. $p = 10^{-4}$).

Apparently the population of the combined areas of Chepigana and Pinogana cannot be considered as homogeneous, hence an analysis based on the crude proportions seropositives in the total sample may produce biased results.

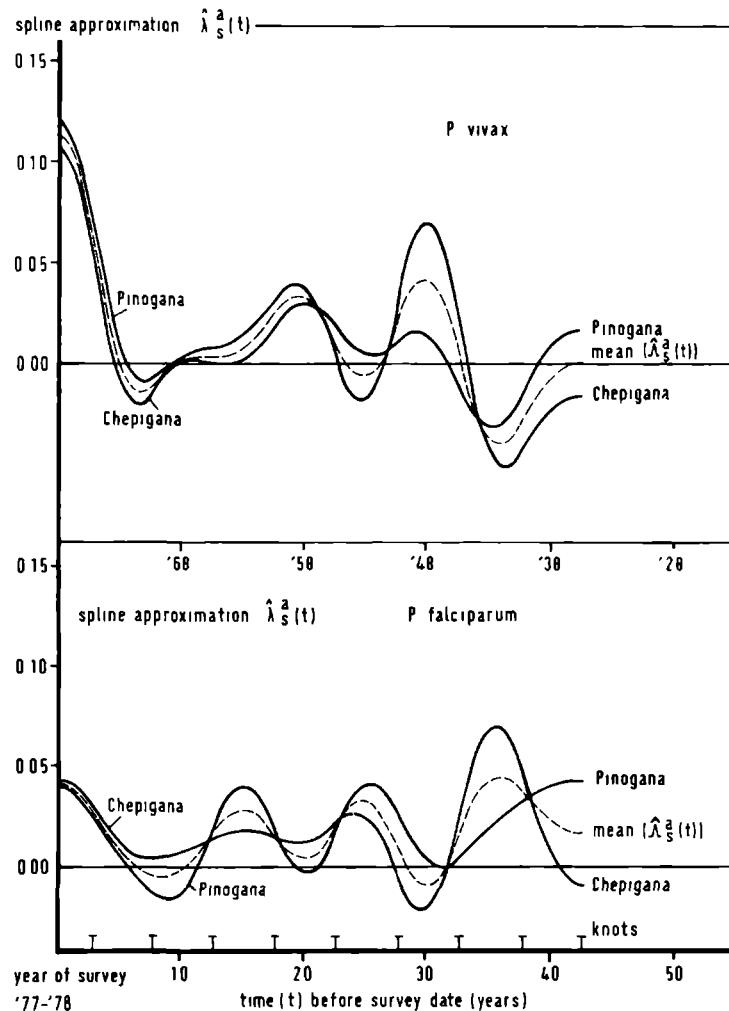
The spline approximations $\hat{\lambda}_s^a(t)$ of Chepigana and Pinogana with their mean $\bar{\lambda}_s^a(t)$, based on the seroreactivity to *P.vivax* and *P.falciparum*, are shown in fig. 5.6.3. For each time point t before survey date and each antigen the following two hypothesis are tested (for a description of these tests see chapter 3, section 3.2):

- Chepigana and Pinogana have equal values $\lambda_s^a(t)$
- Chepigana and Pinogana have equal rates of change $\gamma_s^a(t)$.

A difference in $\lambda_s^a(t)$ is established in period 2.7-3.3 yr before survey date (*P.vivax*) and period 7.1-10.0 yr before survey date (*P.falciparum*); however, no difference in rate of change of $\gamma_s^a(t)$ between Chepigana and Pinogana is found (two-sided test $p \leq 0.05$).

In chapter 3, section 3.3.3, three methods (tests) have been introduced for combining and analysing serological data sampled from a non-homogeneous

Figure 5.6.3 Spline approximations $\hat{\lambda}_s^a(t)$ in Chepigana and Pinogana with their mean $\bar{\lambda}_s^a(t)$.

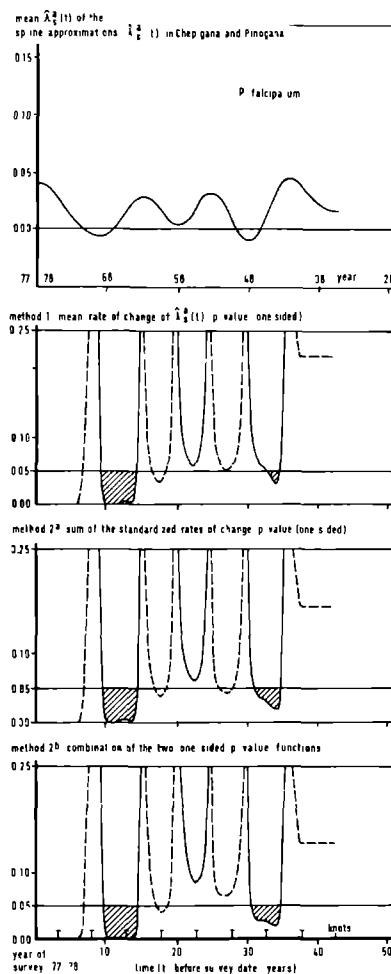
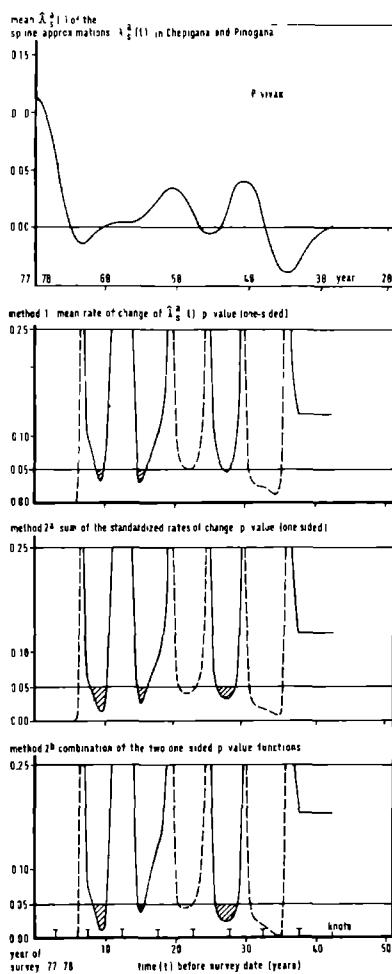


legenda: $\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

population. These methods are applied to the data of table 5.6.1 and table 5.6.2 in order to detect periods of change in the overall trend of the malaria intensity. The results of this analysis are presented in fig. 5.6.4 (separately for the IFA. P.vivax and the IFA. P.falciparum data). Method 1 is based on statistical inference with respect to the mean rate of change of $\hat{\lambda}_s^a(t)$ in Chepigana and Pinogana; method 2^a uses the sum of the standardized rates of change of $\hat{\lambda}_s^a(t)$ and method 2^b combines the one-sided p-value functions of Chepigana and Pinogana as presented in fig. 5.6.1 and fig. 5.6.2. The results of the application of method 1 are analysed further taking into account the factor of past-sensitivity.

The mean $\hat{\lambda}_s^a(t)$ of the spline approximations $\hat{\lambda}_s^a(t)$ in Chepigana and Pinogana based on the seroreactivity to P.vivax shows 3 periods of decrease (fig. 5.6.4, method 1, one-sided p-value, $p \leq 0.05$). With respect to the seroreactivity to P.falciparum antigen, 2 periods of decrease of $\hat{\lambda}_s^a(t)$ are found (fig. 5.6.4, method 1, one-sided p-value, $p \leq 0.05$). These periods are presented in table 5.6.3 (see page 217) (overall analysis step 1). For each time point t in these periods it is verified whether the rate of change $\hat{\lambda}_s^a(t)$ of the overall trend $\hat{\lambda}_s^a(t)$ is larger than the estimate of the formal upperbound $\hat{U}_s(t)$ (for a description of $\hat{U}_s(t)$, see chapter 4, section 4.5.4). Furthermore the maximal linear correction method (MLC) for the factor past-sensitivity is applied separate for Chepigana and Pinogana ($\hat{\gamma}_s^a(t) > \hat{u}_s(t)$). These results are also presented in table 5.6.3. A downward trend of the transmission intensity is indicated for practically the complete periods found in step 1 of the procedure.

Figure 5.6.4 Three methods to combine evidence of change in the spline approximations $\hat{\lambda}_s^a(t)$ in Chepigana and Pirogana.



legenda: $\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

5.6.4 Trend based on seroreactivity to *P.vivax* and *P.falciparum*

The mean $\hat{\lambda}_s^a(t)$ of the spline approximations $\hat{\lambda}_s^a(t)$ of Chepigana and Pinogana based on the seroreactivity to *P.vivax* antigen and *P.falciparum* antigen in the IFA test are shown in fig. 5.6.5 (see page 218). The curves related to the seroreactivity to *P.vivax* and *P.falciparum* show a periodicity with a wavelength of about 10 years and are opposite in phase.

5.6.5 Discussion

If the serological surveys in Chepigana and Pinogana have been carried out in a comparable way then it would appear that the trend of transmission has been different in these two adjacent areas. The serological profiles of Chepigana and Pinogana indicate that the total population cannot be considered as homogeneous. Statistically the difference in seroreactivity mainly concerns the age groups 5-9 yr and 10-14 yr. In Chepigana, as compared with Pinogana, the seroreactivity in these two age groups is lower with respect to *P.vivax* antigen and higher with respect to *P.falciparum* antigen (table 5.6.1 and table 5.6.2). This difference in seroreactivity is reflected in a difference in the level of the spline approximations $\hat{\lambda}_s^a(t)$ in the two populations (see fig. 5.6.3). The level of $\hat{\lambda}_s^a(t)$ related to the seroreactivity to *P.vivax* is significantly lower in Chepigana in period 2.7-3.3 yr before survey date. A lower *P.vivax* transmission intensity in Chepigana in or near to this period may, at least to some extent, explain the lower seropositivity rates in Chepigana in the age groups 5-9 yr and 10-14 yr. The level of $\hat{\lambda}_s^a(t)$ related to the seroreactivity to *P.falciparum* appears to be higher ($p \leq 0.05$) in Chepigana in period 7.1-10.0 yr before survey date; this finding is related to the observation that the *P.falciparum* seropositivity rates in the age groups 5-9 yr and 10-14 yr are higher in Chepigana as compared with Pinogana.

Table 5.6.3 Detection of a downward trend of the transmission intensity in Chepigana and Pinogana; overall analysis, application of the two-step procedure.

Periods before survey date (yr)			
Step 1		Step 2	
Decrease $\hat{\lambda}_s^a(t)$	Classification into subperiods		
$p \leq 0.05$	Overall	Chepigana	Pinogana
IFA P.vivax	$\hat{\gamma}_s^a(t) > \hat{U}_s(t)$	$\hat{\gamma}_s^a(t) > \hat{u}_s(t)$	$\hat{\gamma}_s^a(t) > \hat{u}_s(t)$
8.9 - 9.9	9.6 - 9.9	9.6 - 9.9	9.6 - 9.9
14.7 - 15.8	14.7 - 15.8	14.7 - 15.8	14.7 - 15.8
26.9 - 27.8	26.9 - 27.8	26.9 - 27.8	26.9 - 27.8
IFA P.falciparum	$\hat{\gamma}_s^a(t) > \hat{U}_s(t)$	$\hat{\gamma}_s^a(t) > \hat{u}_s(t)$	$\hat{\gamma}_s^a(t) > \hat{u}_s(t)$
9.4 - 14.4	10.4 - 14.3	9.4 - 14.2	11.3 - 14.1
33.1 - 34.8	33.1 - 34.8	33.1 - 34.7	33.1 - 34.8

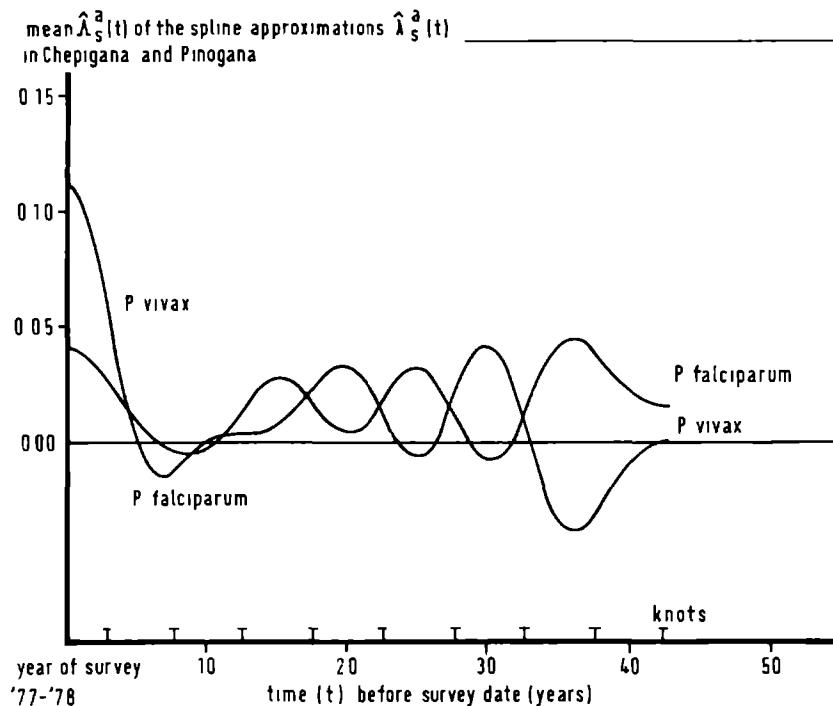
legenda: Only those time points are considered for which $\hat{\lambda}_s^a(t)$ and $\hat{\gamma}_s^a(t)$ respectively assume positive values.

$\hat{\lambda}_s^a(t)$ = mean of $\hat{\lambda}_s^a(t)$, $\hat{\gamma}_s^a(t)$ and $\hat{\gamma}_s^a(t)$ denote the rates of change of $\hat{\lambda}_s^a(t)$ and $\hat{\lambda}_s^a(t)$, $\hat{U}_s(t)$ and $\hat{u}_s(t)$ are upperbounds.

Since there is evidence for a difference in seroreactivity between Chepigana and Pinogana the serological information obtained from the two areas has to be pooled in an appropriate way. In chapter 3, section 3.3.2, it was shown than an analysis based on the crude proportions seropositives might produce misleading results and therefore 3 other methods of combining and analysing serological data were introduced in that chapter. The results of the 3 methods applied to the Panama data show close resemblance (see the curves in fig. 5.6.4). Occasionally method 2^a and method 2^b may have a greater ability to detect changes in the spline approximations $\lambda_s^a(t)$ as seen in this application. Method 1, however has been proposed as the main alternative to

the crude method (see chapter 3, section 3.3.3). In the following the analysis has been continued using the results of this latter method. Correction for the factor of past-sensitivity in the combined areas of Chepigana and Pinogana seems to indicate (see table 5.6.3) a downward trend in *P.vivax* transmission in 3 periods and a downward trend in *P.falciparum* transmission in 2 periods (9.6-9.9, 14.7-15.8, 26.9-27.8 yr before survey date, and 10.4-14.3, 33.1-34.8 yr respectively before survey date).

*Figure 5.6.5 Mean of the spline approximations $\hat{\lambda}_s^a(t)$ in Chepigana and Pinogana based on the seroreactivity to *P.vivax* antigen and *P.falciparum* antigen (IFA test).*



legenda: $\lambda_s^a(t)$ = spline-trend of the antibody related transmission function

Application of the two-step procedure for a homogeneous population to the crude proportions seropositives of the total sample also yields about the same picture of the overall trend in transmission i.e. a downward trend in transmission is indicated for *P.vivax* and *P.falciparum* in the periods 9.6-10.3, 14.5-16.1, 26.2-28.9 yr before survey date and 9.6-14.0, 31.6-34.9 yr before survey date respectively. Apparently the indicated difference in transmission intensity in Chepigana and Pinogana and the difference in sample sizes of the surveys carried out in these two areas do not produce truly different results if we apply the "homogeneous" model.

Unfortunately no information on the malaria history in the two areas is available which could confirm (or disprove) these results. The continuation of this discussion is for that reason rather hypothetical.

Of great interest are the cyclic patterns in the spline trends $\hat{\lambda}_s^a(t)$. Before discussing these results in relation to the biological factors which might cause such patters a few comments are made on the methodological aspects of spline approximation.

The choice of the knots of the spline approximation $\hat{\lambda}_s^a(t)$ determines the type of trend under study. In this application the knots were placed at time points 3.0, 7.5, 12.5, 17.5, 22.5, 27.5, 32.5, 37.5 and 42.5 yr before survey date. In period 7.5-42.5 yr before survey date the knots are placed at an equal distance of 5 years; thus the spline approximations describe 5 yearly trends. The property of preservation of surface of spline approximation (see chapter 2, section 2.1.5) states that the surface of the area below the spline approximation $\hat{\lambda}_s^a(t)$ between two adjacent knots is exactly equal to the corresponding surface of the true but unknown antibody related transmission function $\lambda^a(t)$. Accordingly choosing the knots in other time points on the time axis will lead to another picture of the trend in transmission intensity. This does not imply that the methodology is inconsistent, it simply means that

another type of trend is being studied.

The place of the knots in this application is particularly appropriate to detect a cyclic pattern with a periodicity of 10 years and extreme values in the trend of the transmission intensity in time points 10.0, 15.0, 20.0, 25.0, 30.0, 35.0, 40.0 yr before survey date. The knots, however, were not placed by design to detect this special type of trend. On the contrary the curves presented in fig. 5.6.5 were the surprising results of the application of the spatial and time-dependent transmission model to the seropositivity rates presented in table 5.6.1 and 5.6.2. Without doubt these curves are biased in particular immediately before survey date and again at the end. However the ultimate impression is that the cyclic patterns reflect some reality. The curve related to the seroreactivity to *P.falciparum* antigen shows peaks at or near to the time points 35, 25, 15 yr before survey date and troughs near to the time points 30, 20, 10 yr before survey date.

Extrapolation of this cyclic pattern to the period 0-10 yr before survey date suggest a peak at 5 yr before survey date and minimum transmission intensity on the day of the survey. Continuation of the cyclic pattern in this way is not found in the curve presented in fig. 5.6.5; from this point of view the increase in the *P.falciparum* curve immediately before survey date might be an artefact. Extrapolation of the cyclic pattern of the curve related to the seroreactivity to *P.vivax* in period 12.5-32.5 yr before survey date to the period 0-12.5 yr before survey date would indicate a peak at survey date and 10 years before survey date and furthermore a minimum transmission intensity about 5 years before survey date. It would be very interesting to know if these results can be substantiated further by the use of other sources of information such as malaria surveillance data of Chepigana and Pinogana, if these would be available.

The periodicity and the opposite phase in the spline trends $\hat{\lambda}_s^a(t)$

obtained with *P.falciparum* and *P.vivax* antigens are the unforeseen results of the application of the model. This alternating pattern of the curves is unexpected taking into account the well known - albeit incomplete - serological cross-reactivity of malarious sera in serological tests with *P.falciparum* and *P.vivax* antigen. Periodicity and opposite phase could be due to the method of sampling, the classification of the examined individuals into age groups and the effect of fading of antibodies. However in the remaining part of this discussion, it is assumed that these two phenomena, see fig. 5.6.5, are not mere artefacts.

The relationship between cross-sectional serological data and parasitological findings in a period before survey date is in this study indicated by the mathematical relation between the transmission function $\lambda(t)$ and the antibody related transmission function $\lambda^a(t)$. The transmission function $\lambda(t)$ describes the time-dependent risk of receiving an inoculation from the local mosquito population. By definition an inoculation produces a parasitaemia in a previously uninfected individual. Based on the formal mathematical relation between $\lambda(t)$ and $\lambda^a(t)$ it will be tentatively assumed in the following that the spline approximations $\hat{\lambda}_s^a(t)$ in fig. 5.6.5 are reflecting, to some extent, periodicity and opposite phase in the trend of the inoculation intensity of *P.vivax* and *P.falciparum*. In order to attempt to explain these phenomena some relevant literature is considered first.

It is known from various endemic malarious areas, especially in Asia and the New World, that the prevalence of malaria fluctuates in a regular cycle, varying from five to twenty years in length, depending on the location. *MacDonald (1957)*, discussing these cyclical changes, remarks that the extreme cases of periodicity occur in areas where malaria is notoriously *unstable*. Periodic changes are explained as follows:

"A stimulus, such as malaria infection, produces a reactive restraining

mechanism i.e. immune effector mechanisms. If the reaction does not occur simultaneously with the stimulus, true equilibrium cannot be established and some degree of periodic change is inevitable. Stimulus and reaction are separated by a time which appears to be about two months in vivax malaria, and longer in falciparum malaria. The stimulus, being temporarily unrestrained possess the degree at which equilibrium would be established if the reaction were simultaneous, and the immunity ultimately developed is commensurate with this excessive stimulus, more than sufficient to restrain infection to the natural point of equilibrium. In this way a see-saw motion is set up, infection passing the median point, resistance depressing it below that point, then dilution of resistance, and again exaggerated stimulus to its production. In some degree this must happen in malaria of all types, but mathematical analysis shows that the tendency is likely to be very much more marked in association with unstable malaria". "A tendency to oscillation or periodic movement must therefore be included in the characteristics of the unstable form". (*Macdonald, 1957*).

All the factors deciding for stability or instability concern the local vectors and the environmental conditions. *Macdonald (1957)* and *Pampana (1963)* have listed these factors and the characteristics of the two types. Unstable malaria is based on the presence of a vector which either infrequently takes a blood meal on man or which has a short life expectancy or a combination of both. Under these circumstances the density of the vector has to be high in order to ensure transmission (in the order of 1-10 bites per person per night). Seasonal changes have a very pronounced influence on the intensity of transmission, and the degree of immunity is very variable, being generally low except after epidemics. Seasonal as well as non-seasonal periodicity in malaria transmission relates primarily to fluctuations in the vectorial

capacity¹⁾. An increase in malaria prevalence can be due to a potentiation of the vectorial capacity by a combination of factors which promote the density of the malaria vectors, their longevity and the successful completion of the sporogonic cycle of the parasite in the vectors.

Other alternative explanations for periodicity are presented in the book by Bailey (1971), who discusses several mathematical models for recurrent epidemics of infectious diseases, which theoretically explain that certain patterns of public health intervention tending to control a disease, as well as the entering of new cases in a community may induce periodic waves in disease prevalence. If new cases of the disease are permitted to enter the community a fresh outbreak will occur as soon as the population of susceptibles has increased sufficiently.

Major advances in the development of stochastic models explaining oscillation in disease prevalence are due to Bartlett (1956). This work is primarily related to measles so far as practical interpretations are concerned. A theoretical model for the periodicity of endemic malaria - which is analogous to that for measles described by Bartlett (1956) - has been proposed by Radcliffe (1974). The proposed model disregards various factors that codetermine the intensity of malaria transmission i.a. the development of immunity during successive infections. It indicates that gradual recovery of infected hosts and their subsequent reinfection can theoretically explain the periodic behaviour of the disease.

¹⁾ As in Garrett-Jones (1964) this index is defined "As the number of bites on man that those vectors having bitten an individual on day "t" distribute after the extrinsic cycle of duration "n" during the rest of their life". It can be mathematically expressed by the formula $ma^2 p^n / (-e \log p)$, where "m" is the number of vectors per human individual, "a" represents the man-biting habit and "p" the probability of daily survival of the vector.

Based on mathematical modelling May and Anderson (1973) discuss general biological mechanisms that can generate nonseasonal patterns of disease prevalence. In particular seasonal variation in transmission rates may produce complicated nonseasonal cycles in the prevalence of infection. Yorke *et al.* (1973) and Dietz (1976) have argued that this is responsible for the regular biennial cycle for measles in New York City between 1948 and 1964.

With regard to the phenomenon of opposite phase, seasonal alternations in the prevalence of *P.falciparum* and *P.malariae* parasitaemias in children above the age of 5 years have been observed in the highly endemic malarious area of Garki in Nigeria (Molineaux and Gramiccia, 1980). This alternating pattern was explained by the concept of interference of *P.malariae* by *P.falciparum* due to heterologous and/or nonspecific immune effector mechanisms. These authors suggest that suppression of *P.malariae* parasitaemias by those of *P.falciparum* is of epidemiological importance.

In the present study it is difficult to indicate to what extent the above mentioned factors might have caused the periodicity in the spline-trends of the antibody related transmission functions of Chepigana and Pinogana in Panama. The effect of migration cannot be excluded. People coming from Columbia to Chepigana might have changed intermittently the transmission intensity in that area. Pinogana, however, is said to have a more stable population. Since it shows the same periodicity as Chepigana, migration might not be the central factor for the explanation of the periodic behaviour of the curves.

On the basis of existing knowledge it would be unsatisfactory to say that the periodicity and opposite phase could be due to hypothetical long-term genetic changes in the parasite and/or similar mechanisms as described by Molineaux *et al.* (1977, 1980). With respect to the possible existence of heterologous immunity, the literature shows rather conflicting views (Cohen,

(1973)). Furthermore since Panama is not a hyperendemic area and *P.vivax* is involved instead of *P.malariae* it would be speculative to argue that in Panama an alternation of *P.falciparum* and *P.vivax* might be due to an hypothetical immunological mechanism as described by Molineaux *et al* (1977, 1980).

Long-term parasitological and/or entomological data from Panama are not available. Therefore all explanations will be highly questionable. In the following, however, it will be argued that the features of stable and unstable malaria might to some extent clarify the issue.

The region of Panama is tropical and temperature would permit transmission in most parts of it throughout much of the year. The principal vector is *A. albimanus*; this vector is associated with the presence of unstable malaria (*Panpana* (1963)). It is aided as a vector by other species: in many coastal areas by *A. aquasalis* - in some localities anthropophilous and house frequenting, in others zoophilous and silvan - and by *A. punctimacula* - a sylvan, zoophilous species which, however, will attack man readily - (Lane, 1949). Hence the characteristics of the local vector population are such that a tendency of periodic waves would be possible.

It is a well known fact that *vivax* malaria predominates under unstable conditions and *falciparum* malaria where they are stable. With regard to stability, malaria transmission throughout the world ranges as a long series between the extremes of very high stability, as in equatorial Africa, and the extremely unstable form occurring in Northern India and Sri Lanka. Panama might be classified in between these two extreme types. In the Central American Zone malaria is generally moderate in incidence, rarely reaching the hyperendemic levels common in Africa and it is moderately unstable in type. There are great variations in incidence from place to place. In most of this zone *P.vivax* predominates, *P. falciparum* is secondary to it

(MacDonald, 1957). The hypothesis is brought forward that circumstances in Panama could lead periodically to relatively stable malaria - *P.falciparum* predominant - and relatively unstable malaria - *P.vivax* predominant. This would explain not only the periodic waves in the transmission of malaria but even the typical alternating curves found in this study with regard to *P.vivax* and *P.falciparum*.

5.7 SUMMARY

Based on the models developed in chapter 2, 3 and 4, serological data from Guyana, Surinam, Sri Lanka, Nigeria and Panama are (re-)analysed in order to establish periods in which a decrease in the trend of transmission has occurred.

The re-analysis of the Guyana data, reported by *Lobel et al.* (1976), indicates a downward trend of the transmission intensity in the years 1946-1955 in four sectors in the interior of Guyana. It is concluded that the present model provides a more detailed picture of the trend in transmission than the constant infection rate model of *Draper et al.* (1972).

In Surinam two cross-sectional surveys were carried out with an interval of one year, during August and September 1973 and in the same period of 1974 (*v.d. Kaay*, 1975). The 1973 survey indicates a downward trend of transmission in Alalaparoe (Central Jungle Area) in the years 1964-1966 and analysis of the 1974 data provides evidence for a decrease in transmission in the years 1965-1968. It was concluded that the two-step procedure for the detection of a downward trend of transmission applied to the 1973 and 1974 data produces reasonably consistent results. Although the impression exists that between 1965 and 1967 the inhabitants of Alalaparoe suffered much from malaria, more than in the subsequent years, surveillance data are not available to confirm

these results.

A study has been undertaken in order to assess the trend of transmission in the Kilinochchi district of Sri Lanka. The overall analysis of the serological data collected in 8 sub-areas of the district in Sept-Oct 1978 indicates a downward trend of the transmission intensity which starts after the middle of 1977. In view of the development of DDT resistance to the local vector this insecticide was replaced in the Kilinochchi area by malathion in Sept-Oct 1977 i.e. one year before the date of the serological survey. The results of the analysis - which are supported by the results of the retrospective parasitological study (chapter 3) - are considered to provide evidence for the effectiveness of the substitute insecticide in this area.

Serological data of the Garki research project on the epidemiology and control of malaria, were made available by Dr. L. Molineaux - Malaria Action Programme WHO - for a preliminary analysis. The analysis of data collected before intervention and thereafter, in the previously protected and unprotected populations suggest that the methodology might also be of use for malaria control in highly endemic areas. A more refined analysis is needed, however, to allow a final judgement on the applicability of the model in areas of intense transmission.

Finally, data from Chepigana and Pinogana, two adjacent areas in Panama were placed at our disposal by Dr. A. Nàjera - Pan American Health Organization -. The IFA test was applied in order to determine in both populations the seroreactivity to *P.vivax* and *P.falciparum* antigen. The results of the analysis are unforeseen. The curves related to the seroreactivity to *P.vivax* and *P.falciparum* show a periodicity with a wavelength of about 10 years and are opposite in phase. This alternating pattern was not expected taking into account the well known - albeit incomplete -

serological cross-reactivity in malarious sera. If these cyclic patterns are indeed reflecting alternations in the trend of the inoculation intensity of *P.vivax* and *P.falciparum*, then it is suggested that circumstances in Panama could lead periodically to relatively stable malaria - *P.falciparum* predominant - and relatively unstable malaria - *P.vivax* dominant.

GENERAL DISCUSSION

6

In the following some general remarks are made regarding the applicability of the present malaria model in relation to mathematical approaches by others. Furthermore its applicability to other fields of research will also be indicated.

6.1 THE PRESENT MALARIA MODEL IN RELATION TO OTHER MATHEMATICAL APPROACHES

In recent years malaria has once again become a major health problem. Its transmission resumed in many areas where the disease had been brought under control during the period after the second world war. To permit optimal use of available resources in antimalaria programmes, there remains a need for an efficient method to assess the trend of transmission and to evaluate the results of malaria control programmes. This study indicates that mathematical

analysis of one cross-sectional survey - based on the models proposed in chapter 2, 3 and 4 - may provide adequate information to augment the traditional surveillance and evaluation methods.

The value of mathematical models in the epidemiology and control of malaria has always been an object of controversial discussion. The following two quotations cited by Dietz at the International Biometric Conference Hannover (1970) will underline this. The first is from *S. Peller* (1967): "The mathematical approach to malaria, although tried by marioologists of the rank of Ross and Martini, has been a complete failure. The models have neither theoretical value ... nor are they good for practical purposes. Although they are far from including all factors that have a bearing, they are too clumsy to be of any help". The second quotation is from *Pampana* (1969) "In future when malaria eradication is achieved in Africa, it will be due, first of all, to the computer!"

It has to be admitted that, despite the immense contribution of several research workers during the past 70 years on the quantitative aspects of malaria transmission, mathematical models are not widely used for the design and follow up of eradication and control programs (*Bruce-Chwatt*, 1970). This is not surprising; the data collected during a malaria control programme relate simultaneously to the mutual interactions of three different populations of living creatures: man, anopheline vector and malaria parasite. Moreover each of these three is liable to affected by known and unknown environmental factors and to the effect of control operations. The data therefore concern highly variable populations and the study of malaria is complex in the extreme (*Swaroop*, 1966).

Mathematical models for malaria mainly concern entomological, parasitological and serological data. In the following an indication is given of some basic problems with regard to the collection, interpretation and

modelling of such malariometric data.

The vectorial capacity (*Garrett-Jones, 1961*) before malaria control, during the implementation of antimalaria measures and after their withdrawal is an important indicator of the trend of transmission in endemic areas. The sampling of entomological data, however, generally presents considerable practical difficulties. Moreover entomological parameters are far from constant and subject to wide variation in time and space. Consequently the estimation of the vectorial capacity is generally subject to large errors. Another related key parameter for the transmission of malaria is the basic reproduction rate (*Macdonald, 1957*). In non-mathematical terms this is the number of secondary infections distributed in a community of non-immunes as the direct result of the presence of a single, primary case. The control of malaria aims at the reduction of the basic reproduction rate to a value below one. This rate can be obtained from the vectorial capacity by multiplying this last value by a factor b/r . The factor b denotes the proportion of mosquitoes, having sporozoites in their salivary glands that are actually infectious and $1/r$ is the total number of days during which the infected person maintains fertile gametocytes in his blood. The factor b , and to a minor extent $1/r$, varies considerably. Values of the factor b have been found to range in *nature* from 0.01 to 1.00 (*Macdonald, 1957*) and cannot be readily assessed in the field (*Bruce-Chwatt, 1976*). Hence the basic and ruling reproduction rates are subject to even larger errors than the vectorial capacity.

Models based on age related parasite rates deal to some extent with similar difficulties. The detectability of parasites in the blood is dependent on the parasite density - which relates to the immunity level in the individual - and on the sensitivity of the detection method. Therefore, generally there is a marked difference between the observed proportion

positives and the true proportion positives; moreover this difference varies with age and with the degree of the endemicity of the area under study.

Consequently, major problems in modelling the transmission of malaria based on parasitological data are the time and age dependency of the parameters involved and the changing immunity in man. Moreover, as already indicated, most factors which codetermine the transmission intensity interact and the quantitative aspects varies with each plasmodial species.

An important factor involved in the interpretation of age related parasitological data is the phenomenon that two or more broods of organisms may flourish side by side in the same host; the existence of infection is no barrier to superinfection. This "super infection problem" is reviewed by Fine (1971) and he refers to the models of Macdonald-Irwin (Macdonald, 1950) and Walton-Dietz (presented by Fine (1975)). Both these models ignore the time-dependency of the infection rate and the age-dependency of the recovery rate.

In 1974 another attempt was made by Dietz, Molineaux and Thomas in order to account for the factor of immunity. Their model of the transmission of *P.falciparum* was based on the vectorial capacity (input) and on the age related parasite rates (output). The conclusion was drawn that the model simulates the epidemiology of *P.falciparum* infections with acceptable realism and that it could be used both for planning of malaria control and for teaching the epidemiology and control of malaria. With respect to errors involved in the estimation of the vectorial capacity and of the parasite rates to which the model outputs are compared, Molineaux and Gramiccia (1980) remark that "Unbiased estimates are and may remain impossible to obtain, but a model is epidemiologically satisfactory if it predicts reliably the relationship between variables estimated in a standardized way, even if this way is biased".

A mathematical model for the transmission of malaria using serological data faces in a sense the same problems. There has been a general reluctance to apply in the field of malaria the existing mathematical models based on serological data (e.g. the models of Muench (1959)). This is due to the fact that up till now they have not fulfilled the expectations of the mid sixties, when new serological techniques such as IFA became available. The model of Draper *et al* (1972) for analysing cross-sectional serological data does not take into account a variable transmission intensity and the factor fading of antibodies. This is what the present model attempts to do.

The non-stationary Poisson process has been used to describe the risk of receiving inoculations from the local mosquito population and the first derivative $\lambda_s(t)$ of the natural cubic spline approximation of the mean value function of this process has been used as a measure of the trend of transmission. It is emphasized that the position of the knots of the spline approximation determines the type of trend which is analysed. Furthermore it should be noted that the spline-trend $\lambda_s(t)$ may assume negative values. These features might be considered as disadvantageous when using the spline-trend $\lambda_s(t)$ as a measure of the trend of transmission. However, with regard to the effect of the position of the knots on the interpretation of $\lambda_s(t)$, this feature illustrates the great flexibility of $\lambda_s(t)$ to study in principle all types of trends. Accordingly this property can also be considered as an advantage. With respect to the negative values $\lambda_s(t)$ may assume, this is a clear disadvantage. However, since the objective of this study is the detection of *changes in the trend of transmission* "smoothness and minimum change" of $\lambda_s(t)$ are the main desirable properties for modelling trend functions. And it has been shown in Appendix 2, theorem 2.1, that $\lambda_s(t)$ meets this objective.

With regard to the incorporation into the model of the factor of fading

of antibodies it has to be remarked that it is difficult to obtain quantitative information about it, especially since it is directly dependent again on the transmission intensity. For this reason an indirect correction method for this factor and for the factor of sensitivity of the serological test has been developed, while assuming that the specificity of the serological test is 1. Further research will be needed to establish the validity of this approach.

There is a marked difference between the objectives of the present model and all the previous ones (entomological, parasitological as well as serological); it attempts to assess the trend of transmission using data of one cross-sectional survey. This implies that the assumption of a homogeneous population e.g. that there is no age preference with regard to the receipt of an inoculation - is highly essential in the model constructed here. It is emphasized that this model is largely based on mathematical concepts which aim to come as close as possible to reality, but that no real phenomenon can be expected to be exactly in accordance with it. It is very difficult, if not impossible, to isolate all the biological mechanisms involved and to account mathematically for all of them. The key assumptions of the model have varying degree of plausibility in the different applications. The extent to which other factors need to be incorporated is finally a matter for empirical investigation.

Further studies should concern the possibility of an age related variability in the risk of receiving mosquito bites and the assessment of the past-sensitivity of the serological test. If estimates of the past-sensitivity would become available, then the *maximum linear correction method* (*MLC*) for this factor proposed in chapter 4 might be replaced by a direct correction method. This might finally result in an "unbiased" application of the time-dependent and spatial-dependent transmission models developed in

chapter 2 and chapter 3. Furthermore it is suggested that the basic elements of the methodology presented in this study might be of use for the analysis of parasitological data. It is considered of great potential value to explore this possibility in the future.

This study was undertaken in the hope that one cross-sectional serological survey would give an indication of the trend of transmission and that a wider understanding of the relationship between transmission intensity and serological data would become known. This achievement would be of practical epidemiological importance for the control of malaria especially when the prevalence of malaria is too low to be evaluated reliably by blood slide examination alone, especially in areas where antimalaria drugs are used (Meuwissen, 1974). It is still too early to draw a final conclusion on the applicability of the models developed in chapter 2, 3 and 4. However, based on the analyses of data supplied, it is felt that this approach has laid a sound basis for a new interpretation of cross-sectional serological data and that it ultimately can be used to assess the trend of malaria transmission in areas with different degrees of malaria endemicity.

6.2 APPLICABILITY OF THE MODEL TO OTHER FIELDS OF RESEARCH

In chapter 2, section 2.4.1, it has been argued that the time-dependent transmission model might be used for the epidemiological analysis of diseases other than malaria. In principle, the methodology described in chapter 2 can be applied to any field of research which involves numerical and/or statistical inference on the parameter $\lambda(t)$ of a catalytic differential equation (with appropriate account for errors involved in the measurements). This class of models could be called *spline models for catalytic differential equations*. Each choice of the knots provides a function $\lambda_s(t)$ which measures

a spline-trend of the catalytic force involved.

It is very interesting to note that the *force of infection* of an infectious disease, the *hazard rate* in failure time studies and the *intensity function* of a non-stationary Poisson process can be rendered by catalytic differential equations. These equations occur in epidemiology, in biomedical research, in physics, in industrial reliability problems and many other fields. Some applications of the spline model $\lambda_s(t)$ in epidemiology and biomedical research are briefly indicated in the following.

The hazard function, also known as the instantaneous failure rate, death rate or conditional mortality rate, is a measure of the proneness to failure as a function of time t and it gives the risk of failure per unit of time during the aging process in survival studies. From the above it is indicated that the spline model $\lambda_s(t)$ might be especially applied in animal survival experiments.

Choosing a theoretical distribution in survival studies is often a very difficult task. The exponential, the Weibull and the lognormal distribution and many other functions are used in biomedical research and epidemiology (see for instance Lee (1980)). It is commonly accepted by research workers to classify survival data analysis into two broad classes: parametric if the distribution of the survival times is known to be of a special type and non-parametric if this distribution is unknown. A spline model $\lambda_s(t)$, however, combines the advantages of these two approaches, it can be considered parametric as well as non-parametric. Parametric in the sense that the hazard function consists of polynomials of degree 2 and non-parametric in the sense that the distribution function of survival times does not have to be of a special type. Hence the important feature of a spline model $\lambda_s(t)$ is, that it can be used as a model for any continuous hazard rate $\lambda(t)$ and consequently that it can be applied to study survival in each of the commonly used model

functions indicated above. It is suggested that spline models $\lambda_s(t)$ might be used first in order to select, if possible, a particular type of distribution for the survival data under study. If it is not possible to select an appropriate distribution then the analysis can be continued using the hazard rate $\lambda_s(t)$ and the methodology developed in chapter 2.

Crucial questions in survival analysis often concern the comparison of the hazard rates of an experimental and a control group. In such a situation the "trend value" and "trend-change" test for two homogeneous populations described in chapter 3 might be of use. Survival data from a non-homogeneous population might be analysed with the "spatial-time dependent transmission model"; the stratification to the factor area can be replaced by some other stratification based on one or more factors. To what extent the maximum linear correction (MLC) method proposed in chapter 4 takes into account censoring mechanisms in survival data remains again a matter of further research.

In the past the study of tropical diseases has paved the way for several new developments in the field of medicine. In a similar way it is hoped - in all modesty - that this study on the epidemiology of malaria will prove to be of significance beyond its own specific field.

APPENDIX 1

EPIDEMIOLOGICAL PARAMETERS AND NOTATIONAL CONVENTIONS

1.1 PARAMETERS FOR THE DESCRIPTION OF THE TREND IN TRANSMISSION INTENSITY

An *inoculation* is the receipt of a quantity of sporozoites from the local mosquito population which is sufficiently infectious to induce a parasitaemia in a previously uninfected individual.

Homogeneous and non-homogeneous populations

In this study a population is considered homogeneous if each individual of that population is exposed to the same inoculation risk; this risk may be time-dependent. In the non-homogeneous population the spatial variation in inoculation risk is taken into account. The population under study is divided into m subpopulations Π_i ($i=1,\dots,m$) living in m sub-areas. Each subpopulation is assumed to be homogeneous. In the following the underscript i refers to sub-

population Π_i . Parameters in a non-homogeneous population are denoted by capital letters to distinguish them from parameters in a homogeneous population. Furthermore in homogeneous as well as in non-homogeneous populations, spline approximations of parameters are denoted by the underscript "s".

Parameters in a homogeneous population

$\lambda(t)$ = transmission function
= transmission intensity t years before survey date
= mean number of inoculations per annum inflicted on one individual
 t years before survey date
 $\gamma(t)$ = rate of change of $\lambda(t)$
 $\lambda_s(t)$ = spline approximation of $\lambda(t)$
= spline-trend of the transmission function
 $\gamma_s(t)$ = rate of change of $\lambda_s(t)$

Parameters in a non-homogeneous population

$\Lambda(t)$ = average transmission function
= (weighted¹) mean of the transmission functions in the subpopulations
 Π_i ($i=1, \dots, m$)
 $\Gamma(t)$ = rate of change of $\Lambda(t)$
 $\Lambda_s(t)$ = spline approximation of $\Lambda(t)$
= spline-trend of the average transmission function
 $\Gamma_s(t)$ = rate of change of $\Lambda_s(t)$
 $\sigma^2(\lambda_s(t))$ = (weighted) spatial variance of the trend-values $\lambda_s(t)$ in the sub-
 populations Π_i ($i=1, \dots, m$)
 $\sigma^2(\gamma_s(t))$ = (weighted) spatial variance of the rates of change $\gamma_s(t)$ in the
 subpopulations Π_i ($i=1, \dots, m$)

¹⁾ Weighting factors are for instance the population sizes of the subpopulations
 Π_i ($i=1, \dots, m$).

Age related parameters in a homogeneous population

- $p(A)$ = cumulative inoculation rate
 = proportion of individuals of age A who have experienced at least one infection with micro-organism m in their life¹⁾.
- $p^a(A)$ = seropositivity rate
 = proportion of individuals of age A who are serologically positive - as measured by the test - at the moment of blood collection.
- $\theta(A)$ = S-1 ratio
 = ratio of the seropositivity rate $p^a(A)$ and the cumulative inoculation rate $p(A)$.
- $\eta^a(A)$ = sensitivity of the serological test
 = proportion of individuals of age A who are positive in the test amongst those individuals who have antibodies specific for micro-organism m at the moment of blood collection.
- $\xi^a(A)$ = specificity of the serological test
 = proportion of individuals of age A who are negative in the test amongst those individuals who do not have antibodies specific for micro-organism m at the moment of blood collection.
- $\eta(A)$ = past-sensitivity of the serological test
 = proportion of individuals of age A who are positive in the test amongst those individuals who have experienced at least one infection with micro-organism m in their life.
- $\xi(A)$ = past-specificity of the serological test
 = proportion of individuals of age A who are negative in the test amongst those individuals who never have experienced an infection with micro-organism m in their life.
- $f(A)$ = degree of fading of antibodies
 = proportion of individuals of age A who do not have antibodies specific for micro-organism m at the moment of blood collection

¹⁾ In this study an infection with micro-organism m is equated with malaria.

amongst those individuals who have experienced at least one infection with the micro-organism m in their life.

- $\theta_c(A)$ = critical S-I ratio
 $\eta_c(A)$ = critical past-sensitivity

Age related parameters in a non-homogeneous population

- $P(A)$ = average cumulative inoculation rate
= the complement of the (weighted) geometric mean of $1-p_i^a(A)$ in the subpopulations Π_i ($i=1,\dots,m$).
 $P^a(A)$ = average seropositivity rate
= the complement of the (weighted) geometric mean of $1-p_i^a(A)$ in the subpopulations Π_i ($i=1,\dots,m$).
 $O(A)$ = average S-I ratio
= ratio of the average seropositivity rate $P^a(A)$ and the average cumulative inoculation rate $P(A)$.
 $H(A)$ = average past-sensitivity of the serological test
 $\theta_c^a(A)$ = average critical S-I ratio
 $\eta_c^a(A)$ = average critical past-sensitivity

1.3 ANTIBODY RELATED TRANSMISSION FUNCTIONS

The transmission function $\lambda(t)$ and its spline approximation $\lambda_s(t)$ are derived from the cumulative inoculation rate $p(A)$. If the factors fading of antibodies, the sensitivity and the specificity of the serological test are disregarded, then the seropositivity rate $p^a(A)$ is erroneously equated to the cumulative inoculation rate $p(A)$. Hence the "transmission function" derived from $p^a(A)$ instead of $p(A)$ is "a biased" transmission function. This biased transmission function is called antibody related transmission function, written as $\lambda^a(t)$,

$$\lambda^a(t) = \text{antibody related transmission function}$$

= "transmission function" based on $p^a(A)$ instead of $p(A)$.

Accordingly the notation $\Lambda^a(t)$ is used to indicate that this function is based on the seropositivity rates $p^a(A)$ in the subpopulations Σ_i ($i=1,\dots,m$).

$\Lambda^a(t)$ = average antibody related transmission function

The spline approximations of $\lambda^a(t)$ and $\Lambda^a(t)$ are indicated by $\lambda_s^a(t)$ and $\Lambda_s^a(t)$. The rates of change of these functions in a time point t before survey date are notated as $\gamma^a(t)$, $\Gamma^a(t)$, $\gamma_s^a(t)$ and $\Gamma_s^a(t)$ respectively.

Table A.1.1 and table A.1.2 (see page 244) provide a summary of the parameters defined in a homogeneous population and in a non-homogeneous population.

Table A.1.1 Homogeneous population; parameters derived from the cumulative inoculation rate $p(A)$ and the seropositivity rate $p^a(A)$.

cumulative inoculation rate $p(A)$	seropositivity rate $p^a(A)$
$v(A) = -e \log(1-p(A))$	$v^a(A) = -e \log(1-p^a(A))$
$\lambda(A) = \frac{dv}{dA}$	$\lambda^a(A) = \frac{dv^a}{dA}$
$\gamma(A) = \frac{d\lambda}{dA}$	$\gamma^a(A) = \frac{d\lambda^a}{dA}$
$\theta(A) = \frac{p^a(A)}{p(A)}$	

Spline approximations ¹⁾	
$p_s(A) = 1-e^{-v_s(A)}$	$p_s^a(A) = 1-e^{-v_s^a(A)}$
$\lambda_s(A) = \frac{dv_s}{dA}$	$\lambda_s^a(A) = \frac{dv_s^a}{dA}$
$\gamma_s(A) = \frac{d\lambda_s}{dA}$	$\gamma_s^a(A) = \frac{d\lambda_s^a}{dA}$
$\theta_s(A) = \frac{p_s^a(A)}{p_s(A)}$	

¹⁾ $v_s(A)$ and $v_s^a(A)$ are natural cubic spline approximations of $v(A)$ and $v^a(A)$ respectively.

Table A.1.2 Non-homogeneous population; parameters¹⁾ derived from the cumulative inoculation rates $p_i^a(A)$ and the seropositivity rates $p_i^a(A)$ in the m subpopulations Π_i ($i=1, \dots, m$).

cumulative inoculation rates $p_i^a(A)$		seropositivity rates $p_i^a(A)$	
$P(A) = 1 - \prod(1 - p_i^a(A))$	w_i	$p_i^a(A) = 1 - \prod(1 - p_i^a(A))$	w_i
$\Lambda(A) = \sum w_i \lambda_i^a(A)$		$\Lambda^a(A) = \sum w_i \lambda_i^a(A)$	
$\Gamma(A) = \frac{d\Lambda}{dA}$		$\Gamma^a(A) = \frac{d\Lambda^a}{dA}$	
$\Theta(A) = \frac{p_i^a(A)}{P(A)}$			

Spline approximations

$P_s^a(A) = 1 - \prod(1 - p_{1,s}^a(A))$	w_1	$p_s^a(A) = 1 - \prod(1 - p_{1,s}^a(A))$	w_1
$\Lambda_s^a(A) = \sum w_1 \lambda_{1,s}^a(A)$		$\Lambda_s^a(A) = \sum w_1 \lambda_{1,s}^a(A)$	
$\Gamma_s^a(A) = \frac{d\Lambda_s^a}{dA}$		$\Gamma_s^a(A) = \frac{d\Lambda_s^a}{dA}$	
$\Theta_s^a(A) = \frac{p_s^a(A)}{P_s^a(A)}$			

1) Parameters in a non-homogeneous population are denoted by capital letters to distinguish them from parameters in a homogeneous population.

The functions $v_s(A)$ and $v_s^a(A)$ are natural cubic spline approximations of $v(A)$ and $v^a(A)$ respectively. More comprehensive definitions of the parameters involved in this study, in particular the definitions of the spline approximations of the parameters are presented in Appendix 2, 3 and 4.

Conventions regarding the notation of estimates

In general no distinction is made between the notation of estimates and estimators of parameters. Estimates (estimators) of population parameters are denoted by a "hat" above the corresponding parameter e.g.

$\lambda_s(t)$ = spline approximation of $\lambda(t)$

$\hat{\lambda}_s(t)$ = estimate (estimator) of $\lambda_s(t)$

$\sigma^2(\lambda_s(t))$ = spatial variance in $\lambda_s(t)$

$\hat{\sigma}^2(\lambda_s(t))$ = estimate(estimator) of $\sigma^2(\lambda_s(t))$.

The estimate (estimator) of the standard deviation of an estimator () is denoted by SD(), for example

$SD(\hat{\lambda}_s(t))$ = estimate (estimator) of stand. dev. of $\hat{\lambda}_s(t)$.

Where it may add to the clarity of the notation the parameter t (time before survey date) and the underscript s (spline approximation) is suppressed.

A brief summary of this simplified notation is given in the table below.

Notational conventions

comprehensive notation

simplified notation

parameter	estimate	stand.dev.	parameter	estimate	stand.dev.
$\lambda(t)$			λ	$\hat{\lambda}$	$SD\hat{\lambda}$
$\Lambda(t)$			Λ	$\hat{\Lambda}$	$SD\hat{\Lambda}$
$\lambda_s(t)$	$\hat{\lambda}_s(t)$	$SD(\hat{\lambda}_s(t))$	λ	$\hat{\lambda}$	$SD\hat{\lambda}$
$\Lambda_s(t)$	$\hat{\Lambda}_s(t)$	$SD(\hat{\Lambda}_s(t))$	Λ	$\hat{\Lambda}$	$SD\hat{\Lambda}$
$\sigma^2(\lambda_s(t))$	$\hat{\sigma}^2(\lambda_s(t))$	1)	σ^2_λ	$\hat{\sigma}^2_\lambda$	1)

1) Expressions and symbols for the estimated standard deviation of $\hat{\sigma}^2(\lambda_s(t))$ are not used in this study.

In case we are dealing with several subpopulations then these subpopulations and the corresponding parameters are indicated by the underscript i or j; for instance $\lambda_{i,s}(t)$ is the spline approximation $\lambda_s(t)$ in subpopulation Γ_i . In correspondence with the conventions described above $\lambda_{i,s}(t)$ is often indicated by λ_i .

Estimators of $\Lambda_s(t)$ and $\sigma^2(\lambda_s(t))$ are derived both in a fixed design and a random design. These estimators (estimates) are indicated by the underscripts F and R respectively, e.g.

$\hat{\Lambda}_F$ = estimate (estimator) of $\Lambda_s(t)$ in an F-design

$\hat{\Lambda}_R$ = estimate (estimator) of $\Lambda_s(t)$ in an R-design

$\hat{\sigma}_{F,\lambda}^2$ = estimate (estimator) of $\sigma^2(\lambda_s(t))$ in an F-design

$\hat{\sigma}_{R,\lambda}^2$ = estimate (estimator) of $\sigma^2(\lambda_s(t))$ in an R-design.

The notational conventions regarding the parameters $\gamma(t)$, $\Gamma(t)$, $\gamma_s(t)$, $\Gamma_s(t)$ and $\sigma^2(\gamma_s(t))$ are completely analogous to those described above.

Conventions regarding the description of time and age

In order to avoid ambiguity in the mathematical description and epidemiological interpretation of the spline approximation (spline-trend) $\lambda_s(t)$, it is emphasized that the symbol t indicates a time point t years before survey date. The epidemiological interpretation of the course of a trend function, however, is in the opposite direction i.e. from the past to the survey date. Consequently the rate of change of $\lambda_s(t)$, denoted by $\gamma_s(t)$, has the following epidemiological interpretation:

" $\gamma_s(t) < 0$ " corresponds with "the spline-trend $\lambda_s(t)$ is increasing"

" $\gamma_s(t) > 0$ " corresponds with "the spline-trend $\lambda_s(t)$ is decreasing".

The symbol A may refer to a time point A years before survey date or to age A (years). From the context in which the symbol A is used it will be clear which interpretation is meant, for instance:

$\lambda(A)$ = value of the transmission function $\lambda(t)$

in a time point A years before survey date

$\eta(A)$ = past-sensitivity for age A.

The course of the age related functions in this study are described and interpreted in the conventional way, e.g. $\frac{d\eta}{dA} > 0$ means $\eta(A)$ increases with age.

1.5 GENERAL LEGENDA FOR THE INTERPRETATION OF THE CURVE OF THE SPLINE

APPROXIMATION $\hat{\lambda}_s^a(t)$

In chapter 4, section 4.2.4, a two-step procedure is proposed for the detection of a downward trend of the transmission intensity in a homogeneous population. In all applications presented in chapter 5 a standard type of presentation is used to render the results of this procedure. The standard figure used to present these results consists of an upper-half and a lower-half. In interpreting this figure four parts of it have to be considered consecutively¹⁾:

1. The graphical representation of the spline approximation $\hat{\lambda}_s^a(t)$ of the antibody related transmission function in the upper-half of the figure.
2. The one-sided p-value function used for the detection of a significant decrease in $\hat{\lambda}_s^a(t)$ in the lower-half of the figure (solid curve). If the

¹⁾ Chapter 4, section 4.3, provides a comprehensive illustration of the application of the two-step procedure using serological data from Mauritius.

solid curve falls below 0.05, then the spline approximation $\lambda_s^a(t)$ is considered to decrease in that particular point of time before survey date (single shaded area in the lower-half of the figure).

3. Those periods in which the spline approximation $\lambda_s^a(t)$ is assumed to decrease are indicated in the upper-half of the figure by the single shaded area //.

It is noted that double shading always implies single shading.

4. Finally the correction for the factor of past-sensitivity is considered (*step 2 of the procedure*). Subperiods of the periods indicated in part 3 are shown in the upper-half of the figure, with a decrease in $\hat{\lambda}_s^a(t)$ sufficiently large to assume a decrease in the spline approximation $\lambda_s^a(t)$. These subperiods are indicated by an *additional shading* in the other direction //.

Further remarks regarding the interpretation of the standard figure:

- a) If the one-sided p-value function presented in the lower-half of the figure does not fall below 0.05, then it is decided that the serological data do not provide evidence for a downward trend of transmission and step 2 of the procedure is not performed.
- b) The main objective is the detection of a downward trend in transmission intensity ($\lambda_s^a(t)$). However, occasionally it is also the intention to discuss the detection of periods in which the trend of the transmission intensity might have increased; therefore the associated one-sided p-value curve (interrupted line) in the lower-half of the figure is used.
- c) In order to avoid an erroneous interpretation of the value of the spline approximation $\hat{\lambda}_s^a(t)$, the description of the unit of the vertical axis is deliberately not presented in the upper-half of the figure.

APPENDIX 2

THE TREND OF TRANSMISSION IN A HOMOGENEOUS POPULATION

2.1 THE TIME-DEPENDENT TRANSMISSION MODEL

2.1.1 *The non-stationary Poisson process*

The mathematical base of the time-dependent transmission model is the non-stationary Poisson process. There are several ways one can give systems of axioms that a stochastic process needs to satisfy in order to be a non-stationary Poisson process (see for instance Parzen (1962), Cox and Lewis (1966), Snyder (1975)). In the present application the basic event is the occurrence of an inoculation with malaria parasites inflicted on an individual of the population. (For the definition of inoculation and further details see chapter 2, section 2.1.2 or Appendix I).

Sufficient conditions for the counting process of inoculations to be a non-stationary Poisson process are given below. These conditions do not

imply the most general Poisson process. For our purpose, however, the conditions are sufficiently general to judge, whether the Poisson counting process is a reasonable model for the description of the risk of inoculations (with malaria parasites or infectious agents of some other infectious disease).

The non-stationary Poisson process is characterized in this study as an integer-valued process with independent increments and unit jumps. Furthermore we assume the existence of a continuous intensity function. This intensity function is used as a measure of the transmission intensity of malaria in a homogeneous population.

Consider an individual of age A on the day of the survey, let t be a time point before survey date ($0 \leq t \leq A$). Let $N(t)$ be the number of inoculations inflicted on the individual in period $[0, t]$ before survey date. $N(t)$ is called the counting function (random variable). The collection of random variables $\{N(t); 0 \leq t \leq A\}$ constitutes a stochastic process. The following assumptions are made on the occurrence of an inoculation: (2.1)

1. Since we begin counting inoculations at time $t=0$, we define $N(0)=0$.

For all choices of time points $t_1 < t_2$ in period $[0, A]$ before survey date it is assumed that $0 < P[N(t_2) - N(t_1) > 0] < 1$. In words: in any interval (no matter how small) there is a positive probability that an inoculation will occur, but it is not certain that it will occur.

2. For any time t , $0 \leq t < A$

$$\lim_{\Delta \downarrow 0} \frac{P[N(t+\Delta) - N(t) \geq 2]}{P[N(t+\Delta) - N(t) = 1]} = 0.$$

In words: in sufficiently small intervals at most one inoculation occurs i.e. it is not possible for inoculations to happen simultaneously.

3. The process $\{N(t); 0 \leq t \leq A\}$ has independent increments i.e. for all choices of time points $t_1 < t_2 < \dots < t_n$ in period $[0, A]$ before survey date the

random variables $N(t_2) - N(t_1)$, $N(t_3) - N(t_2)$, ..., $N(t_n) - N(t_{n-1})$ are independent.

4. There exists a continuous function $\lambda(t)$ for which

$$\lim_{\Delta \rightarrow 0} \frac{P[N(t+\Delta) - N(t) = 1]}{\Delta} = \lambda(t).$$

The function $\lambda(t)$ is called the intensity function of the counting process.

A counting process of inoculations which satisfies the conditions 1, 2, 3 and 4 is a non-stationary Poisson process. For the proof the reader is referred to Parzen (1962, chapter 4, theorem 2A) or Snijder (1975, chapter 2, theorem 2.2.2). It follows that the random variable $N(t)$ has a Poisson distribution with mean value $EN(t)$. The mean value function $EN(t)$ is denoted by $v(t)$. The function $\lambda(t)$, defined in condition 4, is the first derivative of $v(t)$ and is called the intensity function of the non-stationary Poisson process. We call this function transmission function. For a set of axioms which contains weaker conditions and for a discussion of non-continuous mean value functions $v(t)$, the reader may consult Khinchin (1956) and Snyder (1975, theorem 2.2.1).

Until now a transmission function $\lambda(t)$ has been defined only for a single individual of the population. Assuming a homogeneous population implies that all individuals of the population are exposed at time t before survey date to the same risk of receiving an inoculum. So the value of $\lambda(t)$ at time t is equal for all individuals of the population under study. Hence we can define $\lambda(t)$ the transmission function of the population and use this function to measure the time-dependent force of potential infections at time t before survey date. It is convenient (not essential) to assume that $\lambda(t)$ is differentiable; the rate of change of $\lambda(t)$ is denoted by $\gamma(t)$.

3.1.2 Spline approximations

Spline functions are piecewise defined polynomials of degree n. The pieces join in the so called knots and fulfil continuity conditions for the function itself and the first $n-1$ derivatives. Thus a spline function of degree n is a continuous function with $n-1$ continuous derivatives. In the following we shall use the term spline approximation in a wide sense, i.e. any function of spline functions which is used for approximation purposes is called spline approximation. The first derivative of the natural cubic spline approximation of the mean value function $v(t)$ of a non-stationary Poisson process is introduced here to define the trend of the intensity function $\lambda(t)$ of this process. Two properties of this trend function, i.e. the property of *preservation of surface* and the *minimum change property* ensure that this type of trend function is very suitable to describe all kind of changes in the "trend" of the intensity function $\lambda(t)$.

Spline approximations of $v(t)$, $\lambda(t)$ and $\gamma(t)$

Consider a division of the time axis before survey date in k intervals $[0, T_1]$, $[T_1, T_2]$, ..., $[T_{k-1}, T_k]$. The time points $0, T_1, T_2, \dots, T_k$ are called knots. A cubic spline approximation $v_s(t)$ of the mean value function $v(t)$ is a function on $[0, T_k]$ which satisfies:

1. $v_s(t)$ is a polynomial of degree 3 in each interval $[0, T_1]$, $[T_1, T_2]$, ..., $[T_{k-1}, T_k]$
2. $v_s(T_j) = v(T_j)$ $j=0, 1, \dots, k$
3. $\frac{d}{dt}v_s(t)$ and $\frac{d^2}{dt^2}v_s(t)$ are continuous in the knots T_1, T_2, \dots, T_{k-1} .

A polynomial of degree 3 is determined by 4 coefficients, the total number of coefficients of the cubic spline $v_s(t)$ is $4k$. Condition 2 induces $2k$ linear

restrictions on these coefficients, condition 3 specifies $2k - 2$ linear restrictions. The total number of linear independent restrictions is $4k - 2$. Identification can be achieved by imposing two additional restrictions which are usually in the form of end point restrictions. To obtain an important minimum property with regard to the second derivative of $v_s(t)$ the end conditions are put in the form

$$4. \quad \left[\frac{d^2}{dt^2} v_s(t) \right]_{t=0} = \left[\frac{d^2}{dt^2} v_s(t) \right]_{t=T_k} = 0. \quad (2.3)$$

A cubic spline approximation $v_s(t)$ which satisfies condition 4 is called the *natural cubic spline approximation* of $v(t)$.

The trend of the intensity function, notation $\lambda_s(t)$, is defined as the first derivative of the natural cubic spline approximation $v_s(t)$, thus we have

$$\lambda(t) = \frac{d}{dt}v(t) \text{ and } \lambda_s(t) = \frac{d}{dt}v_s(t). \quad (2.4)$$

The rate of change in $\lambda_s(t)$ is notated as $\gamma_s(t)$, i.e. by definition

$$\gamma(t) = \frac{d}{dt}\lambda(t) \text{ and } \gamma_s(t) = \frac{d}{dt}\lambda_s(t).$$

The function $\lambda_s(t)$ is a spline function of degree 2 and is called *spline approximation of the transmission function* $\lambda(t)$. The function $\gamma_s(t)$ is a spline function of degree 1, this function describes the rate of change in the (spline) trend of the transmission intensity.

Theorem 2.1 The trend function $\lambda_s(t)$ has the following properties:

$$1. \quad \int_{T_{j-1}}^{T_j} \lambda_s(t) dt = \int_{T_{j-1}}^{T_j} \lambda(t) dt \quad (j=1, \dots, k), \quad (2.5)$$

where T_0, T_1, \dots, T_k are the knots of the spline approximation $\lambda_s(t)$ and T_0 is defined by $T_0=0$.

This property of the trend function $\lambda_s(t)$ is shortly called *property of preservation of surface*.

2. Let the function $c(t)$ be an arbitrary approximation of the transmission function $\lambda(t)$ in period $[0, T_k]$ before survey date which possesses the property of preservation of surface and has a continuous first derivative, then the following relation holds

$$\int_0^{T_k} \left[\frac{d}{dt} \lambda_s(t) \right]^2 dt \leq \int_0^{T_k} \left[\frac{d}{dt} c(t) \right]^2 dt. \quad (2.6)$$

This property is called *minimum change property of the trend function $\lambda_s(t)$* .

Proof of property 1: As we have for $j=1, \dots, k$

$$\int_{T_{j-1}}^{T_j} \lambda_s(t) dt = \int_{T_{j-1}}^{T_j} \frac{d}{dt} v_s(t) dt = v_s(T_j) - v_s(T_{j-1}), \text{ and in the same way}$$

$$\int_{T_{j-1}}^{T_j} \lambda(t) dt = \int_{T_{j-1}}^{T_j} \frac{d}{dt} v(t) dt = v(T_j) - v(T_{j-1}), \text{ it follows from condition 2 of}$$

the definition of the natural cubic spline approximation that

$$\int_{T_{j-1}}^{T_j} \lambda_s(t) dt = \int_{T_{j-1}}^{T_j} \lambda(t) dt \quad (j=1, \dots, k).$$

Proof of property 2: Let the function $f(t)$ be defined by

$$f(t) = \int_0^t c(u) du \quad 0 \leq t \leq T_k,$$

then $f(t)$ is a function which has a continuous second derivative in $[0, T_k]$

and passes through the points $(T_j, v(T_j))$, $j=0, 1, \dots, k$. This is readily

verified since,

$$f(T_j) = \int_0^{T_j} c(t) dt = \sum_{i=1}^j \int_{T_{i-1}}^{T_i} c(t) dt = \sum_{i=1}^j \int_{T_{i-1}}^{T_i} \lambda(t) dt = \int_0^{T_j} \lambda(t) dt = v(T_j).$$

From a theorem of Holladay¹⁾, (see Ahlberg et al., 1967, page 3), which concerns the basic integral relation for natural cubic splines to other interpolating functions having a continuous second derivative, it follows that the natural cubic spline approximation $v_s(t)$ minimizes the integral

$$\int_0^{T_k} \left[\frac{d^2}{dt^2} f(t) \right]^2 dt.$$

Thus for the natural cubic spline approximation $v_s(t)$ of $v(t)$ we have

$$\int_0^{T_k} \left[\frac{d^2}{dt^2} v_s(t) \right]^2 dt \leq \int_0^{T_k} \left[\frac{d^2}{dt^2} f(t) \right]^2 dt. \text{ Hence it follows immediately that}$$

$$\int_0^{T_k} \left[\frac{d}{dt} \lambda_s(t) \right]^2 dt \leq \int_0^{T_k} \left[\frac{d}{dt} c(t) \right]^2 dt. \text{ This completes the proof of theorem 2.1.}$$

Spline approximation of the cumulative inoculation rate $p(A)$

The cumulative inoculation rate $p(A)$ is defined as the proportion of individuals of age A who have experienced at least one inoculation in their life.

¹⁾Theorem (Holladay): Let $\Delta: a = x_0 < x_1 < \dots < x_n = b$ and a set of real numbers $\{y_i\}$ ($i=0, 1, \dots, n$) be given. Then of all functions $f(x)$ having a continuous second derivative on $[a, b]$ and such that $f(x_i) = y_i$ ($i=0, 1, \dots, n$), the spline function $S_\Delta(f; x)$ with junction points at the x_i and with $S''_\Delta(f; a) = S''_\Delta(f; b) = 0$ minimizes the integral

$$\int_a^b |f''(x)|^2 dx.$$

Hence the following relation holds

$$p(A) = p[N(A) > 0] = 1 - e^{-v(A)}. \quad (2.7)$$

The spline approximation ($p_s(A)$) of the cumulative inoculation rate is accordingly defined by

$$p_s(A) = 1 - e^{-v_s(A)}. \quad (2.8)$$

In the practical applications grouped cumulative inoculation rates $\hat{p}(A_j)$ ($j=1, \dots, k$) are analysed with A_j chosen in the middle of age-group j . The knots of the natural cubic spline approximation $v_s(t)$ are placed in time points before survey date which correspond with these midpoints¹⁾ i.e.

$T_j = A_j$ ($j=0, 1, \dots, k$). In the knots A_j ($j=0, 1, \dots, k$) we then have $v_s(A_j) = v(A_j)$ ($j=0, 1, \dots, k$), see (2.2), and thus

$$p_s(A_j) = p(A_j) \quad (j=0, 1, \dots, k). \quad (2.9)$$

The spline approximation $\lambda_s(t)$ has been defined by $\lambda_s(t) = \frac{d}{dt}v_s(t)$. Hence it follows at once from (2.8) that

$$p_s(A) = 1 - e^{-\int_0^A \lambda_s(t) dt}. \quad (2.10)$$

The natural cubic spline approximation $v_s(A)$ of the mean value function $v(A)$ does not need to be order preserving. Therefore the spline approximation $\lambda_s(t)$ may assume negative values. It may even occur that the spline approximation $p_s(A)$ assumes negative values. If the spline approximation $\lambda_s(t)$ is non-negative ($0 \leq t \leq T_k$), then relation (2.10) shows that $p_s(A)$ can be

¹⁾ $T_0 = A_0 = 0$

considered the cumulative inoculation rate of a hypothetical homogeneous population which has been exposed to a transmission intensity " $\lambda_s(t)$ ". Furthermore it follows from the definition of $\lambda(t)$ and $\lambda_s(t)$ and the relations (2.7) and (2.8) that the value of $\lambda(t)$ and $\lambda_s(t)$ in a time point A years before survey date is given by respectively

$$\lambda(A) = \frac{d}{dA} [-e \log(1 - p(A))], \quad \lambda_s(A) = \frac{d}{dA} [-e \log(1 - p_s(A))]. \quad (2.11)$$

Hence the relation between " $\lambda(t)$ and $p(A)$ " and " $\lambda_s(t)$ and $p_s(A)$ " are formally identical. In appendix 3 (see (3.10)) similar relations are established in a non-homogeneous population.

2.2 STATISTICAL INFERENCE

There are several numerical methods to solve the system of equations defining the natural cubic spline. The method of *Walsh, Ahlberg and Nilson (1967)* seems to have the best computational properties. However we are not primarily interested in efficient numerical methods of calculations (recursive methods). For statistical inference it is of more importance to express the spline approximation $\lambda_s(t)$ and its rate of change $\gamma_s(t)$ as weighted linear combinations of the parameters $v(T_j)$ ($j=0, 1, \dots, k$).

The first part of this section concerns the determination of functions $l_j(t)$ and $c_j(t)$, $j=0, 1, \dots, k$, for which

$$\lambda_s(t) = \sum_{j=0}^k l_j(t) v_j \text{ and } \gamma_s(t) = \sum_{j=0}^k c_j(t) v_j.$$

In the second part of this section estimators are derived of the spline approximation $\lambda_s(t)$ and its rate of change $\gamma_s(t)$. Furthermore a statistical test is proposed for the detection of a change in the spline-trend $\lambda_s(t)$ in a

time point t years before survey date ("trend-change test").

2.2.1 Determination of the functions $\lambda_j(t)$, $v_j(t)$; $j=0, 1, \dots, k$

The spline approximation $\lambda_s(t)$ is the first derivative of the natural cubic spline approximation $v_s(t)$. It is convenient to describe $v_s(t)$ in the interval $[T_{j-1}, T_j]$ in a coordinate system for which the time point zero is situated in knot T_{j-1} . Using condition (1) of the definition of $v_s(t)$, see (2.2), we then have

$$v_s(t) = \alpha_{j0} + \alpha_{j1}d + \alpha_{j2}d^2 + \alpha_{j3}d^3, \quad (2.12)$$

where $T_{j-1} \leq t \leq T_j$ and $d = t - T_{j-1}$. Using matrix notation this can be written as

$$v_s(t) = [1, d, d^2, d^3] \alpha_j \text{ with } \alpha_j = [\alpha_{j0}, \alpha_{j1}, \alpha_{j2}, \alpha_{j3}]'. \quad (2.13)$$

A $(4k) \times (k+1)$ matrix A will be derived (see (2.31)) which relates the vector α of spline coefficients $\alpha = [\alpha_1', \dots, \alpha_j', \dots, \alpha_k']'$, to the vector $v = [v(T_0), v(T_1), \dots, v(T_k)]'$,

$$\alpha = Av. \quad (2.14)$$

This matrix A will be used to express the spline function $\lambda_s(t)$ and its rate of change $\gamma_s(t)$ as weighted linear combinations of the parameters $v(T_j)$ ($j=0, 1, \dots, k$).

The vector α_j is determined by the values $v_s(T_{j-1})$, $v_s(T_j)$ and the slopes $\lambda_s(T_{j-1})$, $\lambda_s(T_j)$ of $v_s(t)$ in the knots T_{j-1} , T_j . The following four equations hold: (2.15)

$$v_s(T_{j-1}) = \alpha_{j0}$$

$$v_s(T_j) = \alpha_{j0} + \alpha_{j1}(T_j - T_{j-1}) + \alpha_{j2}(T_j - T_{j-1})^2 + \alpha_{j3}(T_j - T_{j-1})^3$$

$$\lambda_s(T_{j-1}) = \alpha_{j1}$$

$$\lambda_s(T_j) = \alpha_{j1} + 2\alpha_{j2}(T_j - T_{j-1}) + 3\alpha_{j3}(T_j - T_{j-1})^2.$$

Using condition (2) of the definition of $v_s(t)$, see (2.2), we have

$$v_s(T_j) = v(T_j) \quad (j=0,1,\dots,k).$$

For simplicity of notation $v(T_j)$ is written as v_j , thus¹⁾

$$v_j = v(T_j) = v_s(T_j) \quad (j=0,1,\dots,k). \quad (2.16)$$

Furthermore, since it will add to the clarity of the notation, the underscript "s" in $\lambda_s(T_j)$ is suppressed in the following, i.e.

$$\lambda_j = \lambda_s(T_j) \quad (j=0,1,\dots,k). \quad (2.17)$$

This last notation might cause some confusion as $\lambda_j \neq \lambda(T_j)$. In this section, however, λ_j always refers to $\lambda_s(T_j)$ and not to $\lambda(T_j)$. Therefore there will be no ambiguity in the interpretation of λ_j in the following. The difference between two adjacent knots T_{j-1} , T_j is denoted by Δ_j

$$\Delta_j = T_j - T_{j-1} \quad (j=1,\dots,k). \quad (2.18)$$

From equation (2.15), and using (2.16), (2.17) and (2.18), it follows by matrix inversion that

$$\begin{bmatrix} \alpha_{j0} \\ \alpha_{j1} \\ \alpha_{j2} \\ \alpha_{j3} \end{bmatrix} = \begin{bmatrix} 1 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 \\ \frac{-3}{\Delta_j^2} & \frac{3}{\Delta_j^2} & \frac{-2}{\Delta_j} & \frac{-1}{\Delta_j} \\ \frac{2}{\Delta_j^3} & \frac{-2}{\Delta_j^3} & \frac{1}{\Delta_j^2} & \frac{1}{\Delta_j^2} \end{bmatrix} \begin{bmatrix} v_{j-1} \\ v_j \\ \lambda_{j-1} \\ \lambda_j \end{bmatrix}. \quad (2.19)$$

¹⁾ note, $T_0=0$ and thus $v_0 = v(T_0) = v_s(T_0) = 0$

The symbol λ is used to denote the $(k+1)$ column vector of values of the spline approximation $\lambda_s(t)$ in the knots,

$$\lambda = [\lambda_0, \lambda_1, \dots, \lambda_k]' \text{ where } \lambda_j = \lambda_s(T_j), \quad j=0,1,\dots,k.$$

Using equation (2.19) the vector of cubic spline coefficients α can be expressed in the vector $[v, \lambda]'$. That is a matrix K can be derived from (2.19) such that the following relation holds

$$\alpha = K \begin{bmatrix} v \\ \lambda \end{bmatrix}. \quad (2.20)$$

Matrix K is a $(4k) \times (2k+2)$ matrix which can be partitioned in two matrices K_1 and K_2

$$K = [K_1, K_2], \quad (2.21)$$

where K_1 and K_2 are $(4k) \times (k+1)$ matrices defined on the next page. From (2.20) and (2.21) follows $\alpha = [K_1, K_2] \begin{bmatrix} v \\ \lambda \end{bmatrix}$ or equivalently

$$\alpha = K_1 v + K_2 \lambda. \quad (2.22)$$

The defining equations for the slopes $(\lambda_j, j=0,1,\dots,k)$ of the cubic spline approximation $v_s(t)$ in the knots T_0, T_1, \dots, T_k follow from straightforward but tedious application of the conditions (1), (2) and (3) presented in (2.2). For details see Ahlberg et al. (1967), page 13, formula 2.1.16.

$$\left[\begin{array}{ccc|ccc|ccc} 2 & g_0 & & & \lambda_0 & v_0 \\ w_1 & 2 & g_1 & & \lambda_1 & v_1 \\ w_2 & 2 & g_2 & & \lambda_2 & v_2 \\ \cdot & \cdot & \cdot & & \cdot & \cdot \\ \cdot & \cdot & \cdot & & \cdot & \cdot \\ w_{k-2} & 2 & g_{k-2} & & \lambda_{k-2} & v_{k-2} \\ w_{k-1} & 2 & g_{k-1} & & \lambda_{k-1} & v_{k-1} \\ w_k & 2 & & & \lambda_k & v_k \end{array} \right] = \begin{bmatrix} v_0 \\ v_1 \\ v_2 \\ \vdots \\ \vdots \\ v_{k-2} \\ v_{k-1} \\ v_k \end{bmatrix} \quad (2.23)$$

$$\begin{array}{l}
 K_1 = \left[\begin{array}{cc}
 1 & 0 \\
 0 & 0 \\
 \frac{-3}{\Delta_1^2} & \frac{3}{\Delta_1^2} \\
 \frac{2}{\Delta_1^3} & \frac{-2}{\Delta_1^3} \\
 1 & 0 \\
 0 & 0 \\
 \frac{-3}{\Delta_2^2} & \frac{3}{\Delta_2^2} \\
 \frac{2}{\Delta_2^3} & \frac{-2}{\Delta_2^3} \\
 \cdot & \cdot \\
 \cdot & \cdot \\
 \cdot & \cdot \\
 \cdot & \cdot \\
 1 & 0 \\
 0 & 0 \\
 \frac{-3}{\Delta_k^2} & \frac{3}{\Delta_k^2} \\
 \frac{2}{\Delta_k^3} & \frac{-2}{\Delta_k^3}
 \end{array} \right] \quad K_2 = \left[\begin{array}{cc}
 0 & 0 \\
 1 & 0 \\
 \frac{-2}{\Delta_1} & \frac{-1}{\Delta_1} \\
 \frac{1}{\Delta_1^2} & \frac{1}{\Delta_1^2} \\
 0 & 0 \\
 1 & 0 \\
 \frac{-2}{\Delta_2} & \frac{-1}{\Delta_2} \\
 \frac{1}{\Delta_2^2} & \frac{1}{\Delta_2^2} \\
 \cdot & \cdot \\
 \cdot & \cdot \\
 \cdot & \cdot \\
 \cdot & \cdot \\
 0 & 0 \\
 1 & 0 \\
 \frac{-2}{\Delta_k} & \frac{-1}{\Delta_k} \\
 \frac{1}{\Delta_k^2} & \frac{1}{\Delta_k^2}
 \end{array} \right]
 \end{array}$$

The values w_j and g_j ($j=1, \dots, k-1$) are given by

$$w_j = \frac{\Delta_{j+1}}{\Delta_j + \Delta_{j+1}} \quad \text{and} \quad g_j = 1 - w_j \quad (j=1, \dots, k-1). \quad (2.24)$$

The values v_j ($j=1, \dots, k-1$) on the right side of the equation (2.23) are equal to

$$v_j = 3w_j \frac{v_j - v_{j-1}}{\Delta_j} + 3g_j \frac{v_{j+1} - v_j}{\Delta_{j+1}} \quad (j=1, \dots, k-1). \quad (2.25)$$

The values g_0 , v_0 , w_k and v_k in (2.23) depend on the end condition of the cubic spline approximation. Imposing the end conditions of a natural cubic spline

$$\left[\frac{d}{dt} \lambda_s(t) \right]_{T_0} = \left[\frac{d}{dt} \lambda_s(t) \right]_{T_k} = 0,$$

results in the following equations¹⁾

$$\begin{aligned} g_0 &= 1 & w_k &= 1 \\ v_0 &= 3 \frac{v_1 - v_0}{\Delta_1} & v_k &= 3 \frac{v_k - v_{k-1}}{\Delta_k} . \end{aligned} \quad (2.26)$$

Using (2.23) the vector λ can be eliminated from (2.22). First, however, (2.23) is written in matrix notation,

$$W\lambda = v. \quad (2.27)$$

The matrix W is a $(k+1) \times (k+1)$ matrix defined by the left hand side of (2.23) with $g_0 = 1$ and $w_k = 1$. This matrix depends only on the position of the knots on the time axis. The vector v is defined by $v = [v_0, v_1, \dots, v_k]'$. It is readily verified that the coefficients v_j ($j=1, \dots, k-1$) of the vector v represent three times the slope of a parabola through the plane points

¹⁾ For further details, see Ahlberg et al. (1967, page 13).

$(T_{j-1}, v_{j-1}), (T_j, v_j), (T_{j+1}, v_{j+1})$. The equations (2.25) and (2.26) which relate v_0, v_1, \dots, v_k linearly to v_0, v_1, \dots, v_k are easily expressed in matrix notation. Let L be the $(k+1) \times (k+1)$ matrix derived from (2.25) and (2.26) such that

$$v = Lv. \quad (2.28)$$

From (2.27) and (2.28) it follows that λ can be written as

$$\lambda = W^{-1}Lv. \quad (2.29)$$

Substitution of λ in formula (2.22) by (2.29) results in

$$\alpha = K_1 v + K_2 W^{-1} Lv. \quad (2.30)$$

Hence the objective formulated in (2.14) has been achieved i.e.,

$$\alpha = Av \text{ with } A = K_1 + K_2 W^{-1} L. \quad (2.31)$$

The matrix A is a $4k \times (k+1)$ matrix. The matrices K_1 , K_2 , W and L are simple functions of the position of the knots on the time axis. Matrix W is a $(k+1) \times (k+1)$ matrix. To determine the vector of natural cubic spline coefficients α , the inverse of matrix W has to be calculated.

Equation (2.31) is now used to determine functions $l_i(t)$, $i=0,1,\dots,k$ for which

$$\lambda_s(t) = \sum_{i=0}^k l_i(t) v_i.$$

The spline function $\lambda_s(t)$ is the first derivative of $v_s(t)$. From the description of $v_s(t)$, see (2.12), follows that $\lambda_s(t)$ is described in interval $[T_{j-1}, T_j]$ by

$$\lambda_s(t) = \alpha_{j1} + 2\alpha_{j2}d + 3\alpha_{j3}d^2, \quad (2.32)$$

where $T_{j-1} \leq t \leq T_j$ and $d = t - T_{j-1}$.

Matrix A defined in (2.31) can be partitioned in submatrices A_j ($j=1, \dots, k$) such that $\alpha_j = A_j v$. Let A_j^{mi} ($m=0, 1, 2, 3; i=0, 1, 2, \dots, k$) be the coefficients of the matrix A_j , thus

$$\alpha_j = \begin{bmatrix} \alpha_{j0} \\ \alpha_{j1} \\ \alpha_{j2} \\ \alpha_{j3} \end{bmatrix} = \begin{bmatrix} A_j^{00} & A_j^{01} & \dots & A_j^{0k} \\ A_j^{10} & A_j^{11} & \dots & A_j^{1k} \\ A_j^{20} & A_j^{21} & \dots & A_j^{2k} \\ A_j^{30} & A_j^{31} & \dots & A_j^{3k} \end{bmatrix} \begin{bmatrix} v_0 \\ v_1 \\ v_2 \\ \vdots \\ v_k \end{bmatrix}. \quad (2.33)$$

From (2.32) and (2.33) follows that in interval $[T_{j-1}, T_j]$ the spline function $\lambda_s(t)$ is given by

$$\lambda_s(t) = \sum_{i=0}^k (A_j^{1i} + 2d A_j^{2i} + 3d^2 A_j^{3i}) v_i. \quad (2.34)$$

Using (2.34) the spline function $\lambda_s(t)$ can be expressed as a weighted linear combination of the parameters v_i ($i=0, 1, \dots, k$),

$$\lambda_s(t) = l_0(t)v_0 + l_1(t)v_1 + \dots + l_k(t)v_k, \quad (2.35)$$

where $l_i(t)$, $i=0, 1, \dots, k$, are polynomials of degree 2 defined in interval $[T_{j-1}, T_j]$ by

$$l_i(t) = A_j^{1i} + 2A_j^{2i}(t-T_{j-1}) + 3A_j^{3i}(t-T_{j-1})^2. \quad (2.36)$$

The rate of change in $\lambda_s(t)$, i.e. the function $\gamma_s(t)$, is easily derived from (2.35) and (2.36),

$$\gamma_s(t) = c_0(t)v_0 + c_1(t)v_1 + \dots + c_k(t)v_k, \quad (2.37)$$

where $c_i(t)$, $i=0,1,\dots,k$, are polynomials of degree 1 defined in interval $[T_{j-1}, T_j]$ by

$$c_i(t) = 2A_j^{2i} + 6A_j^{3i}(t-T_{j-1}). \quad (2.38)$$

It is emphasized that the coefficients A_j^{mi} ($j=1,\dots,k$; $m=1,2,3$; $i=0,1,\dots,k$) in formula (2.36) and (2.38) only depend on the position of the knots of the natural cubic spline approximation $v_s(t)$. As $v_0 = v(T_0) = v(0) = 0$, the unknown parameters in (2.35) and (2.37) are v_1, v_2, \dots, v_k . The statistical inference regarding the parameters v_j ($j=1,\dots,k$) and the spline functions $\lambda_s(t)$ and $\gamma_s(t)$ are presented in the next section.

2.2.2 Estimation and testing procedures

The first objective is to derive estimators of the trend in transmission intensity $\lambda_s(t)$ and its rate of change $\gamma_s(t)$. It is assumed that the individuals included in the sample of the cross-sectional survey are classified into k age-groups. Let there be n_j individuals in age group A_j and let A_j be the midpoint of the age interval. Only the situation is considered in which the knots of the spline approximation $\lambda_s(t)$ are placed in time points before survey date which correspond with the midpoints of these age intervals i.e. $T_j = A_j$ ($j=1,\dots,k$).

Let A_{ij} be individual i in age group A_j and let $N_{ij}(t)$ be the counting function of inoculations for this individual A_{ij} ($0 \leq t \leq A_j$). The random variable $N_{ij}(A_j)$ follows a Poisson distribution with mean value $v(A_j) = v_j$. On survey date only partial information on the realisation of the counting function N_{ij} is available. In fact it is supposed that we can only distinguish between:

- i) $N_{ij}(A_j) > 0$
- ii) $N_{ij}(A_j) = 0$.

This binary observation is described by the random variable Y_{ij}

$$Y_{ij} = \begin{cases} 1 & \text{if } N_{ij}(A_j) > 0 \\ 0 & \text{if } N_{ij}(A_j) = 0. \end{cases} \quad (2.39)$$

Y_{ij} follows a Bernoulli distribution with parameter $1-e^{-v_j}$. It is supposed that the random variables Y_{ij} are independent. Therefore we can disregard the individual observations Y_{ij} and work only with the fractions (\hat{p}_j) of individuals of age-group A_j who have experienced at least one inoculation,

$$\hat{p}_j = \sum_i Y_{ij} / n_j \quad (j=1, \dots, k). \quad (2.40)$$

In order to apply the method of maximum likelihood (ML) for the estimation of v_j ($j=1, \dots, k$) and to avoid problems regarding the existence of ML estimates if $\hat{p}_j = 1$, it is supposed that n_j is large and that p_j is not too near to 1. For the present need it is assumed that $p_j \leq 1 - \epsilon$ with ϵ an extremely small but fixed value. Theoretically n_j can be larger than $1/\epsilon$ or equivalently $(n_j - 1)/n_j > 1 - \epsilon$. However ϵ is chosen sufficiently small to ensure that the sample sizes n_j ($j=1, \dots, k$) are smaller than $1/\epsilon$ in all practical applications. The ML estimator of p_j is then a) \hat{p}_j if $\hat{p}_j < 1$ and b) $1 - \epsilon$ if $\hat{p}_j = 1$. The parameter v_j is a function of p_j with a single-valued inverse ($v_j = -e \log(1-p_j)$; $0 \leq p_j \leq 1 - \epsilon$). Therefore, using the invariance property of maximum-likelihood estimators (see for instance Mood, Graybill and Boes (1974)), it follows immediately that for $n_j \leq 1/\epsilon$ the ML estimator of v_j is

$$\hat{v}_j = \begin{cases} -e \log(1-\hat{p}_j) & \text{if } \hat{p}_j < 1 \\ -e \log \epsilon & \text{if } \hat{p}_j = 1. \end{cases} \quad (2.41)$$

For $n_j > 1/\epsilon$ the ML estimate of v_j is a) $-e \log(1-\hat{p}_j)$ if $\hat{p}_j \leq 1-\epsilon$ and b) $-e \log \epsilon$ if $\hat{p}_j > 1-\epsilon$. Asymptotically the ML estimator is normally distributed¹⁾. As $n_j \rightarrow \infty$ the asymptotic mean and variance are (Cox, 1970)

$$v_j \text{ and } (e^j - 1)/n_j. \quad (2.42)$$

The asymptotic variance, written as $\sigma^2(\hat{v}_j)$, can be expressed in p_j

$$\sigma^2(\hat{v}_j) = \frac{p_j}{1-p_j} \frac{1}{n_j}. \quad (2.43)$$

$\sigma^2(\hat{v}_j)$ is consistently estimated by replacing p_j by the ML estimator of p_j .

This estimator is denoted by $SD^2\hat{v}_j$. For $n_j \leq 1/\epsilon$ we have

$$SD^2\hat{v}_j = \begin{cases} \frac{\hat{p}_j}{1-\hat{p}_j} \frac{1}{n_j} & \text{if } \hat{p}_j < 1 \\ \frac{1-\epsilon}{1-(1-\epsilon)} \frac{1}{n_j} & \text{if } \hat{p}_j = 1. \end{cases} \quad (2.44)$$

For large values of the sample size n_j in the age group A_j , the expected value of the ML estimator \hat{v}_j will be close to the parameter value v_j . To demonstrate this the ML estimator \hat{v}_j is written as $\hat{v}_j = \phi(\hat{p}_j)$, where the function ϕ is defined on the closed interval $[0,1]$ by

$$\phi(p) = \begin{cases} -e \log(1-p) & p \leq 1-\epsilon \\ -e \log \epsilon & p \geq 1-\epsilon. \end{cases}$$

¹⁾We shall say that a sequence of random variables $\{X_n\}$, $n = 1, 2, \dots$, is asymptotically normally distributed with mean μ_n and variance σ_n^2 if the cumulative distribution function of $(X_n - \mu_n)/\sigma_n$ converges to the cumulative distribution function of the standardized normal variable.

Let p_j be the parameter value in the interval $0 \leq p_j \leq 1-\varepsilon$; thus $E \hat{p}_j = p_j$ and $\phi(p_j) = v_j$. The function ϕ is continuous at p_j . Hence for any arbitrary small value α there exists an interval $(p_j - \delta, p_j + \delta)$, $\delta \neq 0$, for which $|\phi(p) - \phi(p_j)| \leq \alpha/2$ if $|p - p_j| \leq \delta$. Furthermore as $\phi(p)$ is continuous in the closed interval $[0, 1]$, $\phi(p)$ is bounded. Hence there exists an integer M such that $|\phi(p)| \leq M$ for $0 \leq p \leq 1$. It follows immediately that independent of the sample size n_j the following relation holds

$$|E \phi(\hat{p}_j) - \phi(p_j)| \leq \alpha/2 + M P[|\hat{p}_j - p_j| \geq \delta]. \quad (2.45)$$

Using the Chebyshev inequality it is readily seen that

$$P[|\hat{p}_j - p_j| \geq \delta] \leq \frac{p_j(1-p_j)}{n_j \delta^2}. \quad (2.46)$$

Hence it follows at once from (2.45) and (2.46) that $|E \phi(\hat{p}_j) - \phi(p_j)| \leq \alpha$ for sufficiently large values of n_j . This implies the asymptotic unbiasedness of the ML estimator \hat{v}_j , i.e.

$$E \hat{v}_j \rightarrow v_j \quad (n_j \rightarrow \infty). \quad (2.47)$$

In an analogous way it can be derived that the expected value of $(n_j SD^2 \hat{v}_j)$ comes close to $n_j \sigma^2(\hat{v}_j)$, where $\sigma^2(\hat{v}_j)$ is the *asymptotic variance* of \hat{v}_j defined by (2.43). It is readily seen that (j fixed, $n_j = 1, 2, \dots$)

$$E(n_j SD^2 \hat{v}_j) \rightarrow \frac{p_j}{1-p_j} \quad (n_j \rightarrow \infty).$$

This entails that the quotient of the two sequences $E(SD^2 \hat{v}_j)$ and $\sigma^2(\hat{v}_j)$, where $n_j = 1, 2, \dots$ converges to 1, i.e. that we have the *asymptotic equivalence*¹⁾

¹⁾ We shall say that two sequences $\{a_n\}$ and $\{b_n\}$, $n=1, 2, \dots$ are *asymptotically equivalent* if the quotient a_n/b_n converges to 1 ($n \rightarrow \infty$).

of $E(SD^2\hat{v}_j)$ and $\sigma^2(\hat{v}_j)$, this is written as

$$E(SD^2\hat{v}_j) \sim \sigma^2(\hat{v}_j) \quad (n_j \rightarrow \infty). \quad (2.48)$$

Modified estimators

For n_j small or p_j near to 1 occasionally realisations $\hat{p}_j = 1$ can be expected. The estimates \hat{v}_j and $SD^2\hat{v}_j$ are then expressed in the rather arbitrary value e (see (2.41) and (2.44)). If only because of this, the estimators \hat{v}_j and $SD^2\hat{v}_j$ need some modification. A rather crude way of modifying these estimators is to replace \hat{p}_j by $(n_j - 1)/n_j$ if $\hat{p}_j = 1$. If it is additionally assumed that $p_j \leq 1 - 1/n_j$, then these modified estimators can still be considered ML estimators. A more plausible way, however, of modifying \hat{v}_j is to shift first all proportions \hat{p}_j and then apply the log transformation.

For X having the binomial distribution $B(n, p)$, Haldane (1955) and Anscombe (1956) independently suggested the use of $\log(X + \frac{1}{2})/(n+1)$ to estimate $\log p$. The bias incurred is of the order n^{-2} when n is large. Hence, by symmetry, a simple way of modifying the estimator $-e \log(1 - \hat{p}_j)$ is to use

$$\hat{v}_j = -e \log\left(1 - \frac{n_j \hat{p}_j + \frac{1}{2}}{n_j + 1}\right) \quad (2.49)$$

as an estimator of v_j .

A more preferable estimator from a statistical point of view has been introduced by Haldane (1956) and Cook, Kerridge and Pryce (1974). In his 1956 paper, Haldane conjectured that if X is $B(n, p)$ and if $\tau(X) = D(X) - D(n)$, where $D(\cdot)$ is the digamma function¹⁾ then $E\tau(X) = \log p + o(n^{-k})$ for every positive integer k . Such an estimator was termed "almost unbiased" by Haldane.

¹⁾The digamma and trigamma functions are defined by $D(m) = \frac{d}{dm} \log m!$ and $T(m) = \frac{d^2}{dm^2} \log m!$, $m \geq 0$.

In 1974 this conjecture was verified for $0 < p \leq 1$ by Cook et al. Using the digamma function $D(\cdot)$ and the trigamma function $T(\cdot)$ it was shown that if $p > 0$ the estimator $\tau(X) = D(X) - D(n)$ is almost unbiased for $\log p$ and furthermore $\eta(X) = T(X) - T(n)$ is almost unbiased for the variance of $\tau(X)$. Using these results it is easily seen that an almost unbiased estimator of v_j is given by

$$\hat{v}_j = D(n_j) - D(n_j - n_j \hat{p}_j). \quad (2.50)$$

Furthermore $T(n_j - n_j \hat{p}_j) - T(n_j)$ is an almost unbiased estimator of the variance of \hat{v}_j .

Statistical inference on the spline approximation $\lambda_s(t)$

To estimate the value of the spline approximation $\lambda_s(t)$ in a time point t years before survey date the estimator

$$\hat{\lambda}_s(t) = l_1(t) \hat{v}_1 + l_2(t) \hat{v}_2 + \dots + l_k(t) \hat{v}_k \quad (0 \leq t \leq T_k) \quad (2.51)$$

is used. The $\hat{v}_1, \hat{v}_2, \dots, \hat{v}_k$ are the ML estimators of the parameters v_1, v_2, \dots, v_k (see (2.41)). The total sample size in the k age groups is denoted as n , $n = \sum_{j=1}^k n_j$. It is supposed that n_j/n converges to a value c_j as $n \rightarrow \infty$ ($0 < c_j < 1$, $j=1, \dots, k$). The estimator $\hat{\lambda}_s(t)$ is asymptotically normally distributed with mean $\lambda_s(t)$ as $n \rightarrow \infty$. The *asymptotic variance*¹⁾, written as $\sigma^2(\hat{\lambda}_s(t))$, is

$$\sigma^2(\hat{\lambda}_s(t)) = \sum_{j=1}^k l_j(t)^2 \sigma^2(\hat{v}_j), \quad (2.52)$$

where $\sigma^2(\hat{v}_j)$ is the asymptotic variance of \hat{v}_j (see (2.43)).

¹⁾ A clear distinction has to be made between the spatial variance $\sigma^2(\lambda_s(t))$ and the asymptotic variance $\sigma^2(\hat{\lambda}_s(t))$ of the estimator $\hat{\lambda}_s(t)$.

For the sequence of ML estimators \hat{v}_j , where $n_j = 1, 2, \dots$, it has been derived that \hat{v}_j is an asymptotically unbiased estimator of v_j (2.47). Furthermore we have the asymptotic equivalence of the two sequences $E(SD^2\hat{v}_j)$ and $\sigma^2(\hat{v}_j)$ ($n_j \rightarrow \infty$). The asymptotic unbiasedness of $\hat{\lambda}_s(t)$ follows at once from that of \hat{v}_j , thus

$$E \hat{\lambda}_s(t) \rightarrow \lambda_s(t) \quad (n \rightarrow \infty). \quad (2.53)$$

In words: for arbitrary but fixed t the expected value of the estimator $\hat{\lambda}_s(t)$ is close to the parameter value $\lambda_s(t)$ for large sample sizes in the age groups ($j=1, \dots, k$). Another desirable property of a sequence of estimators is consistency. The estimators \hat{v}_j ($j=1, \dots, k$) in (2.51) are maximum likelihood estimators. The consistency of the sequence of estimators $\{\lambda_s(t)\}_n$, where $n \rightarrow \infty$ follows immediately from that of \hat{v}_j ($j=1, \dots, k$).

The asymptotic variance $\sigma^2(\hat{\lambda}_s(t))$ is estimated by $SD^2(\hat{\lambda}_s(t))$,

$$SD^2(\hat{\lambda}_s(t)) = \sum_{j=1}^k l_j(t)^2 SD^2\hat{v}_j, \quad (2.54)$$

where $SD^2\hat{v}_j$ is defined by (2.44). From (2.48) it follows that the quotient of the two sequences $E(SD^2(\hat{\lambda}_s(t)))$ and $\sigma^2(\hat{\lambda}_s(t))$ converges to 1 as $n \rightarrow \infty$, i.e. we have the asymptotic equivalence of these two sequences

$$E(SD^2(\hat{\lambda}_s(t))) \sim \sigma^2(\hat{\lambda}_s(t)) \quad (n \rightarrow \infty). \quad (2.55)$$

The consistency of the estimator $SD^2(\hat{\lambda}_s(t))$ follows from that of $SD^2(\hat{v}_j)$.

The asymptotic properties of $\hat{\lambda}_s(t)$ and $SD(\hat{\lambda}_s(t))$ will be further used in Appendix 3 for the assessment of the spatial variation in the trend of the transmission intensity.

The collection of estimators $\{\hat{\lambda}_s(t), 0 \leq t \leq T_k\}$ constitutes a stochastic process. For large sample sizes n_j ($j=1, \dots, k$) the random variables $\hat{\lambda}_s(t)$ are

approximately normally distributed with mean value function $\lambda_s(t)$ and covariance kernel

$$\sum_{j=1}^k l_j(t_1) l_j(t_2) \sigma^2(\hat{v}_j). \quad (2.56)$$

The correlation coefficient between two estimators $\hat{\lambda}_s(t_1)$, $\hat{\lambda}_s(t_2)$ is estimated by

$$\frac{\sum_{j=1}^k l_j(t_1) l_j(t_2) \frac{\hat{p}_j}{1-\hat{p}_j} \frac{1}{n_j}}{\text{SD } (\hat{\lambda}_s(t_1)) \text{ SD } (\hat{\lambda}_s(t_2))} \quad (\hat{p}_j \neq 1). \quad (2.57)$$

In order to compare the spline-trend $\lambda_s(t)$ at two time points t_1 and t_2 before survey date $\hat{\lambda}_s(t_2) - \hat{\lambda}_s(t_1)$ is treated as an observed value of a normal random variable having mean $\lambda_s(t_2) - \lambda_s(t_1)$ and variance

$$\sum_{j=1}^k (l_j(t_2) - l_j(t_1))^2 \frac{\hat{p}_j}{1-\hat{p}_j} \frac{1}{n_j} \quad (\hat{p}_j \neq 1). \quad (2.58)$$

To test the hypothesis $H_0 : \lambda_s(t_2) = \lambda_s(t_1)$ the two-sided p-value of the corresponding standardized normal variable is calculated.

The estimated transmission function $\hat{\lambda}_s(t)$ has (relative) extrema in the endpoints $t = 0$ and $t = T_k$ and there may exist at most one (relative) extremum in each interval. To examine the differences between the "observed" relative extrema the method described above (see (2.58)) has been applied with $\hat{\lambda}_s(t_1)$ and $\hat{\lambda}_s(t_2)$ two relative extrema of the spline function $\hat{\lambda}_s(t)$. This method has been used in chapter 2 for exploratory purposes and should not be regarded as a "statistical test" in stricto sensu.

Trend-change test for a homogeneous population

To estimate the rate of change ($\gamma_s(t)$) in the trend of the transmission

intensity t years before survey date an analogous way is followed as for the estimation of $\lambda_s(t)$

$$\hat{\gamma}_s(t) = c_1(t) \hat{v}_1 + c_2(t) \hat{v}_2 + \dots c_k(t) \hat{v}_k \quad (0 \leq t \leq T_k). \quad (2.59)$$

The estimate $\hat{\gamma}_s(t)$ is treated as an observed value of a normal random variable having mean $\gamma_s(t)$ and standard deviation

$$SD(\hat{\gamma}_s(t)) = \sqrt{\sum_{j=1}^k c_j(t)^2 \frac{\hat{p}_j}{1 - \hat{p}_j} \frac{1}{n_j}} \quad (\hat{p}_j \neq 1). \quad (2.60)$$

The spline approximation $\hat{\gamma}_s(t)$ consists of piecewise defined polynomials of degree 1. By definition the value of $\hat{\gamma}_s(t)$ is zero in the endpoints $t = 0$ and $t = T_k$ accordingly the standard deviation of $\hat{\gamma}_s(t)$ is zero in these endpoints ($SD \hat{\gamma}_s(0) = SD \hat{\gamma}_s(T_k) = 0$).

To detect a change in the spline trend $\lambda_s(t)$ in a time point t years before survey date ($0 < t < T_k$) the following hypothesis is tested ("trend-change test"):

H_0 : There is no change in the trend of the transmission intensity t years before survey date, $\gamma_s(t) = 0$, against the two-sided alternative
 H_1 : There is a change in the trend of the transmission intensity t years before survey date, $\gamma_s(t) \neq 0$.

The statistic $\hat{\gamma}_s(t)/SD(\hat{\gamma}_s(t))$ is used to test H_0 with $\hat{\gamma}_s(t)$ and $SD(\hat{\gamma}_s(t))$ defined by (2.59) and (2.60). Using the large sample normal approximation of the distribution of this statistic a two-sided p-value is calculated. The asymptotic normality of the test statistic follows from the asymptotic normality of $\hat{\gamma}_s(t)$ and from the consistency of the denominator ($n \rightarrow \infty$).

In order to briefly survey the situation at each individual time point before survey date such a two-sided p-value has been calculated for all t

$(0 < t < T_k)$. This p-value function is plotted. Periods before survey date in which the p-value function falls below 0.05 are considered to indicate periods of change in the trend of transmission ($\gamma_s(t) \neq 0$). The p-value function is a continuous function in period $0 < t < T_k$ before survey date and is only intended to explore the situation at each time point t . In period $0 < t \leq T_1$ and $T_{k-1} \leq t < T_k$ respectively the p-value function takes on a constant value. This is easily derived taking into account the fact that $\hat{\gamma}_s(t)$ is a linear function of time in these periods and that $\hat{\gamma}_s(0) = \hat{\gamma}_s(T_k) = 0$.

The collection of estimators $\{\hat{\gamma}_s(t), 0 < t < T_k\}$ constitutes a stochastic process. For large sample sizes n_j ($j=1, \dots, k$) the estimators $\hat{\gamma}_s(t)$ are approximately normally distributed with mean value function $\gamma_s(t)$ and covariance kernel, given by a formula which is analogous to (2.56). The correlation coefficient between two estimators $\hat{\gamma}_s(t_1)$ and $\hat{\gamma}_s(t_2)$ is estimated using a statistic similar to the statistic in (2.57). The functions $l_j(t)$ are replaced by $c_j(t)$, $j=1, \dots, k$, and $\hat{\lambda}_s(t_1)$, $\hat{\lambda}_s(t_2)$ are substituted by $\hat{\gamma}_s(t_1)$ and $\hat{\gamma}_s(t_2)$ respectively.

It is emphasized that the inference on the change in the trend of the transmission intensity presented here is basically time point related. At a later stage of the research activities we intend to develop a simultaneous statistical procedure for the assessment of $\gamma_s(t)$.

APPENDIX 3

THE TREND OF TRANSMISSION IN A NON-HOMOGENEOUS POPULATION

3.1 SPATIAL-DEPENDENT AND TIME-DEPENDENT TRANSMISSION MODEL

Heterogeneity in a population with respect to the risk of receiving an inoculation may occur within age-classes, between age-classes and between subpopulations living in different areas. In the model presented here the spatial variation in the trend of the transmission intensity is taken into account. The area is divided into m sub-areas and the non-homogeneous population Π is assumed to consist of m homogeneous subpopulations Π_i living in the m sub-areas ($i=1, \dots, m$).

$$\Pi = \bigcup_{i=1}^m \Pi_i, \quad \Pi_i \cap \Pi_j = \emptyset \quad (1 \leq i < j \leq m).$$

In order to define for a non-homogeneous population the average cumulative inoculation rate and the average transmission function weighting factors w_i are used for the subpopulations Π_i ($i=1, \dots, m$). We may choose for instance ($\sum w_i = 1$):

- $w_i = \frac{1}{m}$ (equal weights); or
- = proportionate to the size of subpopulation Π_i ; or
- = proportionate to the size of sub-area i .

For the non-homogeneous population¹⁾ it is natural to define the average transmission function, notation $\Lambda(t)$, by

$$\Lambda(t) = \sum w_i \lambda_i(t). \quad (3.1)$$

The rate of change in $\Lambda(t)$ is denoted by $\Gamma(t)$,

$$\Gamma(t) = \frac{d}{dt} \Lambda(t). \quad (3.2)$$

The definition of the average transmission function $\Lambda(t)$ by (3.1) is the motivation to define the average cumulative inoculation rate, notation $P(A)$, as the cumulative inoculation rate of a hypothetical homogeneous population which has been imaginarily exposed to the average transmission intensity $\Lambda(t)$, i.e. by definition

$$P(A) = 1 - \exp \left[- \int_0^A \Lambda(t) dt \right]. \quad (3.3)$$

The average cumulative inoculation rate $P(A)$ can be easily expressed in the cumulative inoculation rates $p_i(A)$ ($i=1, \dots, m$) in the m subpopulations. It

¹⁾ Parameters for a non-homogeneous population are denoted by capital letters.

follows from (3.1) and (3.3) that

$$P(A) = 1 - \exp \frac{A}{\int_0^{\infty} \sum_i w_i \lambda_i(t) dt} = 1 - \exp \frac{A}{\sum_i w_i \int_0^A \lambda_i(t) dt} = 1 - \prod_i \left(\exp \frac{A}{\int_0^{w_i} \lambda_i(t) dt} \right)^{w_i}.$$

The cumulative inoculation rate $p_i(A)$ in the subpopulation Π_i ($i=1, \dots, m$) is equal to $p_i(A) = 1 - \exp \frac{A}{\int_0^A \lambda_i(t) dt}$. Using the expression for $P(A)$ it follows at once that

$$P(A) = 1 - \prod_i (1 - p_i(A))^{w_i}. \quad (3.4)$$

In words: $1 - P(A)$ is the weighted geometric mean of $1 - p_i(A)$ ($i=1, \dots, m$).

The spline approximation of $\Lambda(t)$, notation $\Lambda_s(t)$, is defined by

$$\Lambda_s(t) = \sum_i w_i \lambda_{i,s}(t). \quad (3.5)$$

The rate of change in $\Lambda_s(t)$ is denoted by $\Gamma_s(t)$,

$$\Gamma_s(t) = \frac{d}{dt} \Lambda_s(t). \quad (3.6)$$

The spline approximation of the average cumulative inoculation rate, written as $P_s(A)$, is defined in correspondence with the definition of $P(A)$

$$P_s(A) = 1 - \exp \frac{A}{\int_0^A \Lambda_s(t) dt}. \quad (3.7)$$

The spline approximation $p_{i,s}(A)$ for subpopulation Π_i ($i=1, \dots, m$) is defined by (see (2.8)) $p_{i,s}(A) = 1 - \exp(-v_{i,s}(A))$. Furthermore $v_{i,s}(A)$ is equal to $\int_0^A \lambda_{i,s}(t) dt$. From these relations and (3.7) it is easily seen that $P_s(A)$ can be expressed in $p_{i,s}(A)$ in a similar way as $P(A)$ is expressed in $p_i(A)$, that is

$$P_s(A) = 1 - \prod_i (1 - p_{i,s}(A))^{w_i}. \quad (3.8)$$

From Appendix 2, formula (2.9) and the formulae (3.4) and (3.8) it follows

that the spline approximation $P_s(A)$ passes through the values of the average cumulative inoculation rate $\Lambda(A)$ in the knots,

$$P_s(A_j) = \Lambda(A_j) \quad (j=0,1,\dots,k). \quad (3.9)$$

The definition of $P_s(A)$ and $\Lambda(A)$ are further justified by the following relations, which follow immediately from the definitions (3.3) and (3.7). In a time point A years before survey date the value of $\Lambda(t)$ and $\Lambda_s(t)$ are equal to

$$\Lambda(A) = \frac{d}{dA} [-e \log(1-P(A))] \text{ and } \Lambda_s(A) = \frac{d}{dA} [-e \log(1-P_s(A))]. \quad (3.10)$$

These relations for a non-homogeneous population are similar to the corresponding relations for a homogeneous population, see Appendix 2, formula (2.11). Furthermore theorem 2.1, which concerns the relationship between the transmission function $\lambda(t)$ and its spline approximation $\lambda_s(t)$ in a homogeneous population, can be generalized to a non-homogeneous population.

Theorem 3.1 The spline approximation $\Lambda_s(t)$ has the following properties:

$$1. \quad \int_{T_{j-1}}^{T_j} \Lambda_s(t) dt = \int_{T_{j-1}}^{T_j} \Lambda(t) dt \quad (j=1,\dots,k), \quad (3.11)$$

where T_j ($j=0,1,\dots,k$) are the knots of the spline approximation. This property of $\Lambda_s(t)$ is called *property of preservation of surface*.

2. Let the function $C(t)$ be an arbitrary approximation of the average transmission function $\Lambda(t)$ in period $[0, T_k]$ before survey date which possesses the property of preservation of surface and has a continuous first derivative, then the following relation holds

$$\int_0^{T_k} \left[\frac{d}{dt} \Lambda_s(t) \right]^2 dt \leq \int_0^{T_k} \left[\frac{d}{dt} C(t) \right]^2 dt. \quad (3.12)$$

This property is called *minimum change property* of the average trend function $\Lambda_s(t)$.

Proof: The property of preservation of surface of $\Lambda_s(t)$ immediately follows from the corresponding property of $\lambda_s(t)$. For T_j ($j=1, \dots, k$) we have

$$\int_{T_{j-1}}^{T_j} \Lambda_s(t) dt = \sum_{i=1}^m \int_{T_{j-1}}^{T_j} \lambda_{i,s}(t) dt = \sum_{i=1}^m \int_{T_{j-1}}^{T_j} \lambda_i(t) dt = \int_{T_{j-1}}^{T_j} \Lambda(t) dt.$$

The value of the transmission function $\lambda(t)$ in a time point A years before survey date is equal to $\lambda(A) = \frac{d}{dA} [-e \log(1-p(A))]$, see (2.11), and $\lambda_s(A)$ has been defined as the first derivative of the natural cubic spline approximation of the mean value function $v(A)$, where $v(A) = -e \log(1-p(A))$ (see (2.4)). Furthermore $\Lambda(A) = \frac{d}{dA} [-e \log(1-P(A))]$ and $\Lambda_s(A) = \frac{d}{dA} [-e \log(1-P_s(A))]$, see (3.10). Therefore if it can be proved that $-e \log(1-P_s(A))$ is the natural cubic spline approximation of $-e \log(1-P(A))$, then $\Lambda_s(A)$ is defined and related to $\Lambda(A)$ in analogous way as $\lambda_s(A)$ is defined and related to $\lambda(A)$. Hence by analogy the minimum change property follows then immediately from theorem 2.1.

From the definition of the average cumulative inoculation rate it follows that $-e \log(1-P(A)) = \sum_{i=1}^m [-e \log(1-p_i(A))] = \sum_{i=1}^m v_i(A)$. The function $v_i(A)$ is the mean value function of the non-stationary Poisson process in subpopulation Π_i ($i=1, \dots, m$). As the natural cubic spline operator is a linear operator it follows that the natural cubic spline approximation of $-e \log(1-P(A))$ is $\sum_{i=1}^m v_{i,s}(A)$. From the definition of $p_{i,s}(A)$, see (2.8), and formula (3.8) it follows that

$$\sum_{i=1}^m v_{i,s}(A) = \sum_{i=1}^m [-e \log(1-p_{i,s}(A))] = -e \log(1-P_s(A)).$$

Hence the natural cubic spline approximation of $-e \log(1-P(A))$ is $-e \log(1-P_s(A))$; this completes the proof of theorem 3.1.

Using the concepts average cumulative inoculation rate $P(A)$ and its spline approximation $P_s(A)$ we may formally replace the non-homogeneous population by a hypothetical homogeneous population with cumulative inoculation rate $P(A)$ and spline approximation $P_s(A)$. The corresponding "homogeneous" transmission function and its spline approximation then represent the average transmission function $\Lambda(t)$ and its spline approximation $\Lambda_s(t)$.

Spatial variation

As a measure of a) the spatial variation in the trend of the transmission intensity and b) the spatial variation in the rate of change therein, the weighted variance of $\lambda_{i,s}(t)$ and $\gamma_{i,s}(t)$ respectively in the m sub-populations Π_i ($i=1, \dots, m$) is used. These variances are denoted by $\sigma^2(\lambda_s(t))$ and $\sigma^2(\gamma_s(t))$,

$$\begin{aligned}\sigma^2(\lambda_s(t)) &= \sum_{i=1}^m w_i [\lambda_{i,s}(t) - \Lambda_s(t)]^2 \\ &= \text{spatial variance in } \lambda_s(t).\end{aligned}\quad (3.13)$$

The formula for $\sigma^2(\gamma_s(t))$ is similar to (3.13), $\lambda_{i,s}(t)$ and $\Lambda_s(t)$ are replaced by $\gamma_{i,s}(t)$ and $\Gamma_s(t)$ ($i=1, \dots, m$).

3.2 STATISTICAL INFERENCE

In this section estimators are derived of the average trend of the transmission intensity $\Lambda_s(t)$, its rate of change $\Gamma_s(t)$ and the spatial variation parameters $\sigma^2(\lambda_s(t))$ and $\sigma^2(\gamma_s(t))$. Furthermore a statistical test is proposed for the detection of a change in the spline-trend $\Lambda_s(t)$ at a time point t years before survey date.

Two sample designs are considered: the *fixed design (F-design)* and the *random design (R-design)*. In an F-design cross-sectional surveys are carried

out in all subpopulations Π_i ($i=1, \dots, m$). In an R-design cross-sectional surveys are only performed in a (weighted) random sample of subpopulations $\Pi_{i_1}, \Pi_{i_2}, \Pi_{i_1}, \Pi_{i_2}, \dots, \Pi_{i_n}$.

As described in Appendix 1, section 1.4, the symbol t (time before survey date) and the underscript s (spline approximation) is often suppressed in the formulae of parameters and their estimators (estimates). For instance the notations λ_i , Λ and σ_λ^2 are used rather than the more complicated notations $\lambda_{i,s}(t)$, $\Lambda_s(t)$ and $\sigma^2(\lambda_s(t))$ respectively. Accordingly the asymptotic unbiasedness of $\hat{\lambda}_s(t)$ with respect to $\lambda_s(t)$ and the asymptotic equivalence of the two sequences $E(SD^2(\hat{\lambda}_s(t)))$ and $\sigma^2(\hat{\lambda}_s(t))$ in subpopulation Π_i , see Appendix 2 formulae (2.53) and (2.55), are briefly written as¹⁾

$$E \hat{\lambda}_i \rightarrow \lambda_i \quad \text{and} \quad E(SD^2 \hat{\lambda}_i) \sim \sigma^2(\hat{\lambda}_i) \quad (N_i \rightarrow \infty). \quad (3.14)$$

In (3.14) the underscript i is fixed and N_i is the total sample size of the k age groups in subpopulation Π_i . The total sample size in the m subpopulations is denoted by N , $N = \sum_{i=1}^m N_i$. It is supposed that N_i/N converges to a value C_i as $N \rightarrow \infty$ ($0 < C_i < 1$, $i=1, \dots, m$).

3.2.1 Fixed design

In an F-design the estimators of the trend function Λ and the spatial variance σ_λ^2 are written as $\hat{\Lambda}_F$ and $\hat{\sigma}_{F,\lambda}^2$. The asymptotic variance of $\hat{\Lambda}_F$ is denoted by $\sigma^2(\hat{\Lambda}_F)$ and its estimator is notated as $SD^2(\hat{\Lambda}_F)$.

To estimate the value of the trend function Λ in a time point t years before survey date the estimator

¹⁾ A clear distinction has to be made between the spatial variance σ_λ^2 and the asymptotic variance $\sigma^2(\hat{\lambda})$ of the estimator $\hat{\lambda}$.

$$\hat{\lambda}_F = \sum_{i=1}^m w_i \hat{\lambda}_i \quad (3.15)$$

is used. It is readily seen that $\hat{\lambda}_F$ is an asymptotically unbiased estimator of Λ . From $E \hat{\lambda}_i \rightarrow \lambda_i$ ($N_i \rightarrow \infty$) it follows immediately that $E \hat{\lambda}_F \rightarrow \Lambda$ ($N \rightarrow \infty$). The consistency of the estimator $\hat{\lambda}_F$ as $N \rightarrow \infty$ follows from the consistency of the estimators $\hat{\lambda}_i$ ($i=1, \dots, m$). The *asymptotic variance* of $\hat{\lambda}_F$, written as $\sigma^2(\hat{\lambda}_F)$, is equal to

$$\sigma^2(\hat{\lambda}_F) = \sum_{i=1}^m w_i^2 \sigma^2(\hat{\lambda}_i), \quad (3.16)$$

where $\sigma^2(\hat{\lambda}_i)$ is the asymptotic variance of $\hat{\lambda}_i$ defined in (2.52). This asymptotic variance $\sigma^2(\hat{\lambda}_F)$ is estimated by

$$SD^2(\hat{\lambda}_F) = \sum_{i=1}^m w_i^2 SD^2(\hat{\lambda}_i), \quad (3.17)$$

where $SD^2(\hat{\lambda}_i)$ is defined in Appendix 2 formula (2.54). From the asymptotic equivalence of $E(SD^2\hat{\lambda}_i)$ and $\sigma^2(\hat{\lambda}_i)$, see (3.14), it follows that the quotient of the two sequences $E(SD^2\hat{\lambda}_F)$ and $\sigma^2(\hat{\lambda}_F)$ converges to 1 as $N \rightarrow \infty$, i.e.

$$E(SD^2\hat{\lambda}_F) \sim \sigma^2(\hat{\lambda}_F) \quad (N \rightarrow \infty). \quad (3.18)$$

To estimate the spatial variance $\sigma_\lambda^2 = \sum_{i=1}^m w_i (\lambda_i - \Lambda)^2$ it seems natural to make use of the statistic $\sum_{i=1}^m w_i (\hat{\lambda}_i - \hat{\lambda}_F)^2$. Generally, however, this would result in an over estimation of the spatial variance. In order to correct for this bias the following estimator is used for the estimation of σ_λ^2

$$\hat{\sigma}_{F,\lambda}^2 = \sum_{i=1}^m w_i (\hat{\lambda}_i - \hat{\lambda}_F)^2 - \sum_{i=1}^m w_i (1-w_i) SD^2\hat{\lambda}_i. \quad (3.19)$$

It is conjectured that $\hat{\sigma}_{F,\lambda}^2$ is an asymptotically unbiased estimator of the spatial variance σ_λ^2 . The exact proof, however, would involve technical

details which are beyond the objectives of this study. Therefore we satisfy ourselves to make plausible that the expected value of $\hat{\sigma}_{F,\lambda}^2$ will be close to σ_λ^2 . This kind of conjectured approximate equality (without explicit mathematical meaning) will be indicated by the symbol \approx . The argument relies on the following approximate equalities ($i=1, \dots, m$): (3.20)

$$\begin{aligned} E(\hat{\lambda}_i) &\approx \lambda_i & E(SD^2\hat{\lambda}_i) &\approx \sigma^2(\hat{\lambda}_i) \\ E(\hat{\lambda}_i - \lambda)^2 &\approx \sigma^2(\hat{\lambda}_i) & E(\hat{\Lambda}_F - \Lambda)^2 &\approx \sigma^2(\hat{\Lambda}_F). \end{aligned}$$

Note that two of these approximate equalities have already been made rigorous (see (3.14)). To demonstrate that $E(\hat{\sigma}_{F,\lambda}^2) \approx \sigma_\lambda^2$, the "total variance" $\sum_{i=1}^m w_i (\hat{\lambda}_i - \hat{\Lambda}_F)^2$ is split up into several components,

$$\begin{aligned} \sum_{i=1}^m w_i (\hat{\lambda}_i - \hat{\Lambda}_F)^2 &= \sum_{i=1}^m w_i (\hat{\lambda}_i - \lambda_i + \lambda_i - \Lambda + \Lambda - \hat{\Lambda}_F)^2 = & (3.21) \\ &= \sum_{i=1}^m w_i (\hat{\lambda}_i - \lambda_i)^2 + \sum_{i=1}^m w_i (\lambda_i - \Lambda)^2 + \sum_{i=1}^m w_i (\Lambda - \hat{\Lambda}_F)^2 + \\ &+ 2 \sum_{i=1}^m w_i (\hat{\lambda}_i - \lambda_i)(\lambda_i - \Lambda) + 2 \sum_{i=1}^m w_i (\hat{\lambda}_i - \lambda_i)(\Lambda - \hat{\Lambda}_F) + \\ &+ 2 \sum_{i=1}^m w_i (\lambda_i - \Lambda)(\Lambda - \hat{\Lambda}_F). \end{aligned}$$

For the expected values of the components on the right hand side of this equation we (approximately) have

$$\begin{cases} E(\sum_{i=1}^m w_i (\hat{\lambda}_i - \lambda_i)^2) \approx \sum_{i=1}^m w_i \sigma^2(\hat{\lambda}_i) \\ E(\sum_{i=1}^m w_i (\lambda_i - \Lambda)^2) = \sigma_\lambda^2 \\ E(\sum_{i=1}^m w_i (\Lambda - \hat{\Lambda}_F)^2) = E(\Lambda - \hat{\Lambda}_F)^2. \end{cases} \quad (3.22)$$

For the expected values of the cross-products in (3.21) it follows that

$$\begin{cases} E(2 \sum_{i=1}^m w_i (\hat{\lambda}_i - \lambda_i)(\lambda_i - \Lambda)) \approx 0 \\ E(2 \sum_{i=1}^m w_i (\hat{\lambda}_i - \lambda_i)(\Lambda - \hat{\Lambda}_F)) = -2E(\Lambda - \hat{\Lambda}_F)^2 \\ E(2 \sum_{i=1}^m w_i (\lambda_i - \Lambda)(\Lambda - \hat{\Lambda}_F)) = 0. \end{cases} \quad (3.23)$$

From (3.21), (3.22) and (3.23) it is easily seen that the expected value of $\sum w_i (\hat{\lambda}_i - \hat{\Lambda}_F)^2$ is approximately equal to

$$\sigma_\lambda^2 + \sum w_i \sigma^2(\hat{\lambda}_i) = E(\Lambda - \hat{\Lambda}_F)^2.$$

As $E(\Lambda - \hat{\Lambda}_F)^2 \sim \sigma^2(\hat{\Lambda}_F)$ and $\sigma^2(\hat{\Lambda}_F) = \sum_{i=1}^m w_i^2 \sigma^2(\hat{\lambda}_i)$, see (3.16) and (3.20), it follows that

$$E(\sum w_i (\hat{\lambda}_i - \hat{\Lambda}_F)^2) \approx \sigma_\lambda^2 + \sum w_i (1 - w_i) \sigma^2(\hat{\lambda}_i),$$

where $\sigma^2(\hat{\lambda}_i)$ is the asymptotic variance of the estimator $\hat{\lambda}_i$ ($i=1, \dots, m$).

From $E(SD^2 \hat{\lambda}_i) \approx \sigma^2(\hat{\lambda}_i)$, see (3.20), it follows that $E(\sum w_i (1 - w_i) SD^2(\hat{\lambda}_i)) \approx \sum w_i (1 - w_i) \sigma^2(\hat{\lambda}_i)$ and thus we have (see 3.19),

$$E(\hat{\sigma}_{F,\lambda}^2) \approx \sigma_\lambda^2. \quad (3.24)$$

For the present need the use of the estimator $\hat{\sigma}_{F,\lambda}^2$ is sufficiently motivated.

Very likely $\hat{\sigma}_{F,\lambda}^2$ is an asymptotically unbiased and consistent estimator of the spatial variance σ_λ^2 .

The variation of the spline approximations $\hat{\lambda}_i$ ($i=1, \dots, m$) at a time point t years before survey date is the result of the influence of two factors: the spatial variation in the trend of the transmission intensity as described by σ_λ^2 and the statistical errors in the estimates $\hat{\lambda}_i$ ($i=1, \dots, m$) due to the sampling of individuals in the subpopulations. These errors are "area and time dependent". In order to study the influence of these two sources of variation on the total variation in $\hat{\lambda}_i$ ($i=1, \dots, m$), the "total variance" is split up into two components called the spatial variance and the error variance. These components are derived from formula (3.19),

$$v_{F,tot} = total\ variance = \sum_{i=1}^m w_i (\hat{\lambda}_i - \hat{\Lambda}_F)^2 \quad (3.25)$$

$$V_{F,\text{spat}} = \text{spatial variance} = \hat{\sigma}_{F,\lambda}^2 \quad (3.26)$$

$$V_{F,\text{err}} = \text{error variance} = \sum_{i=1}^m w_i (1 - w_i) \text{SD}^2 \hat{\lambda}_i. \quad (3.27)$$

Using this convention the total variance $V_{F,\text{tot}}$ can be written as the sum of the spatial variance $V_{F,\text{spat}}$ and the error variance $V_{F,\text{err}}$,

$$V_{F,\text{tot}} = V_{F,\text{spat}} + V_{F,\text{err}}. \quad (3.28)$$

The spatial variance and the error variance are considered to generate the total variance. That part of the total variance that can be explained by statistical errors due to the sampling of individuals is called proportion explained variance¹⁾ and notated as E_F ,

$$E_F = \frac{V_{F,\text{err}}}{V_{F,\text{tot}}}. \quad (3.29)$$

For the estimation of σ_Y^2 formula (3.19) can be used with $\hat{\lambda}_i$ and $\hat{\gamma}_i$ substituted by $\hat{\gamma}_i$ and $\hat{\Gamma}_F$, with $\hat{\Gamma}_F$ defined by $\hat{\Gamma}_F = \sum_{i=1}^m w_i \hat{\gamma}_i$. Furthermore the total variance, the spatial variance, the error variance and the proportion explained variance of $\hat{\gamma}_i$ ($i=1, \dots, m$) are defined completely analogous to the "analysis of variance" concepts used for the description of the variation of $\hat{\lambda}_i$ ($i=1, \dots, m$).

Trend-change test for a non-homogeneous population (fixed design)

By analogy it is readily seen from (3.15) that $\hat{\Gamma}_F = \sum_{i=1}^m w_i \hat{\gamma}_i$ is an asymptotically unbiased and consistent estimator of the rate of change Γ in the trend of the transmission intensity. The *asymptotic variance* of $\hat{\Gamma}_F$,

¹⁾ Note that the proportion explained variance E_F may assume a value > 1 .

written as $\sigma^2(\hat{\gamma}_F)$, is equal to

$$\sigma^2(\hat{\gamma}_F) = \sum_{i=1}^m w_i^2 \sigma^2(\hat{\gamma}_i), \quad (3.30)$$

where $\sigma^2(\hat{\gamma}_i)$ is the asymptotic variance of $\hat{\gamma}_i$ ($i=1, \dots, m$). This asymptotic variance $\sigma^2(\hat{\gamma}_F)$ is estimated by $SD^2(\hat{\gamma}_F) = \sum_{i=1}^m w_i^2 SD^2(\hat{\gamma}_i)$, where $SD^2(\hat{\gamma}_i)$ is defined in Appendix 2, formula (2.60). By analogy, see (3.18), we also have the asymptotic equivalence of the two sequences $E(SD^2(\hat{\gamma}_F))$ and $\sigma^2(\hat{\gamma}_F)$ as $N \rightarrow \infty$. Furthermore as $SD^2\hat{\gamma}_i$ is a consistent estimator of $\sigma^2(\hat{\gamma}_i)$ it follows that $SD^2(\hat{\gamma}_F)$ is consistently estimating the asymptotic variance $\sigma^2(\hat{\gamma}_F)$.

To detect a change in the average spline trend Λ in a time point t years before survey date, the hypothesis $H_0: \Gamma = 0$ is tested against the two-sided alternative $H_1: \Gamma \neq 0$. The same procedure is followed as for a homogeneous population. To test H_0 the statistic

$$T_F(t) = \frac{\hat{\gamma}_F}{SD_F} \quad (3.31)$$

is used. The asymptotic normality of $T_F(t)$ under H_0 follows from the asymptotic normality of $\hat{\gamma}_F$ and from the consistency of the denominator in (3.31).

In order to briefly survey the situation at each individual time point t before survey date, a p-value function, written as $P_F(t)$ has been calculated. Those periods in which $P_F(t)$ falls below 0.05 are considered to reflect a change in the average trend function $\Lambda_s(t)$.

3.2.2 Random design

In a random design, notation R-design, a two-stage sampling procedure is carried out. Let I be a random variable with $P[I=i] = w_i$ ($i=1, \dots, m$). In order to obtain a weighted random sample of subpopulations n independent observations of the random variable I are taken, these observations are denoted

by i_1, i_2, \dots, i_n . The cross-sectional surveys are carried out in the corresponding subpopulations $\Pi_{i_1}, \Pi_{i_2}, \dots, \Pi_{i_n}$. If it occurs that a subpopulation Π_i appears twice or more in this "sample" of subpopulations, then it is supposed that a corresponding number of identical serological surveys are carried out in this subpopulation. The random spline approximation associated with Π_{i_j} and its estimator with estimated standard deviation are written as λ_{i_j} and $\hat{\lambda}_{i_j}$, $SD^2\hat{\lambda}_{i_j}$ respectively ($j=1, \dots, n$)¹⁾.

In the following estimators are derived of the trend function Λ and the spatial variance σ_λ^2 , these estimators are written as $\hat{\Lambda}_R$ and $\hat{\sigma}_{R,\lambda}^2$. The estimator of the variance of $\hat{\Lambda}_R$ is denoted by $SD^2\hat{\Lambda}_R$.

To estimate the value of the trend function Λ in a time point t years before survey date the estimator

$$\hat{\Lambda}_R = \frac{1}{n} \sum_{j=1}^n \hat{\lambda}_{i_j}, \quad (3.32)$$

is used. An unbiased estimator of the variance of $\hat{\Lambda}_R$ is

$$SD^2\hat{\Lambda}_R = \frac{1}{n(n-1)} \sum_{j=1}^n (\hat{\lambda}_{i_j} - \hat{\Lambda}_R)^2. \quad (3.33)$$

To estimate the spatial variance $\sigma_\lambda^2 = \sum_{i=1}^m w_i (\lambda_i - \Lambda)^2$ it seems natural to make use of the statistic $\frac{1}{n-1} \sum_{j=1}^n (\hat{\lambda}_{i_j} - \hat{\Lambda}_R)^2$. However, as $\hat{\lambda}_{i_j}$ is only an estimate of λ_{i_j} ($j=1, \dots, n$), this would generally result in an over estimation of the spatial variance σ_λ^2 . In order to take into account the statistical errors in the estimates $\hat{\lambda}_{i_j}$ ($j=1, \dots, n$), the following estimator is used for the estimation of σ_λ^2

$$\hat{\sigma}_{R,\lambda}^2 = \frac{1}{n-1} \sum_{j=1}^n (\hat{\lambda}_{i_j} - \hat{\Lambda}_R)^2 - \frac{1}{n} \sum_{j=1}^n SD^2\hat{\lambda}_{i_j}. \quad (3.34)$$

¹⁾ The λ_{i_j} , $\hat{\lambda}_{i_j}$ should not be confused with the λ_i and $\hat{\lambda}_i$ in the fixed design: the composed indices refer to the random design.

To demonstrate that for large sample sizes in all age groups the values of the estimates $\hat{\lambda}_R$ and $\hat{\sigma}_{R,\lambda}^2$ are close to the average trend value λ and the spatial variance σ_λ^2 , the non-repeated two-stage sampling procedure ($n=1$) is considered first.

The weighted random selection of one subpopulation Π_i ($i=1, \dots, m$) and corresponding spline approximation λ_i ($i=1, \dots, m$) is described by the following two random variables (*first-stage*),

$I = \text{index } i \text{ of the selected subpopulation } \Pi_i$

$\lambda = \text{spline approximation } \lambda_i \text{ of the selected subpopulation } \Pi_i$.

The possible realisations of I and λ are $\{1, 2, \dots, m\}$ and $\{\lambda_1, \lambda_2, \dots, \lambda_m\}$ respectively. Thus the distributions of the random variables I and λ are given by

$$P[I = i] = P[\lambda = \lambda_i] = w_i \quad (i=1, \dots, m).$$

At the *second stage* of the sampling procedure, given $I = i$, an estimate (with its standard deviation) is obtained of the trend function of the particular subpopulation selected at the first stage. These two values (estimate and estimated standard deviation of the estimator) are considered realisations of two random variables of the combined experiment, they are denoted by $\hat{\lambda}$ and $SD\hat{\lambda}$,

$\hat{\lambda} = \text{estimate of the spline approximation selected at stage one}$

$SD\hat{\lambda} = \text{standard deviation of this estimate.}$

Hence the selection of one subpopulation and the estimation of the trend function in this selected subpopulation is described by the random vector $(I, \lambda, \hat{\lambda}, SD\hat{\lambda})$.

The unconditional expectation and variance of the unobservable random

variable λ is simply given by

$$E\lambda = \sum_{i=1}^m w_i \lambda_i = \Lambda \quad \text{Var } \lambda = \sum_{i=1}^m w_i (\lambda_i - \Lambda)^2. \quad (3.35)$$

In order to assess the unconditional expectation of the observable random variable $\hat{\lambda}$ and $SD^2\hat{\lambda}$ it is convenient to define conditionally on the realization $I = i$ the random variables $\hat{\lambda}_i$ and $SD^2\hat{\lambda}_i$,

$$\hat{\lambda}_i = \hat{\lambda}|I = i \text{ and } SD^2\hat{\lambda}_i = SD^2\hat{\lambda}|I = i. \quad (3.36)$$

Using the relation $E\hat{\lambda} = \sum_{i=1}^m E[\hat{\lambda}_i|I = i] P[I=i]$ and (3.36) the expected value of the random variable $\hat{\lambda}$ is written as

$$E\hat{\lambda} = \sum_{i=1}^m w_i E\hat{\lambda}_i, \quad (3.37)$$

where $E\hat{\lambda}_i = E[\hat{\lambda}|I = i]$. Using the asymptotic unbiasedness of each $\hat{\lambda}_i$ ($i=1, \dots, m$), see (3.14), it follows at once that the unconditional expectation of $\hat{\lambda}$ converges to Λ , i.e.

$$\hat{\lambda} \rightarrow \Lambda \quad (N \rightarrow \infty). \quad (3.38)$$

Furthermore $E(SD^2\hat{\lambda}) = \sum_{i=1}^m w_i E(SD^2\hat{\lambda}_i)$ and for each i ($i=1, \dots, m$) we have the asymptotic equivalence of $E(SD^2\hat{\lambda}_i)$ and $\sigma^2(\hat{\lambda}_i)$ ($N_i \rightarrow \infty$) (see 3.14). Hence it follows that the ratio of the two sequences $E(SD^2\hat{\lambda})$ and $\sum_{i=1}^m w_i \sigma^2(\hat{\lambda}_i)$ converges to 1 as $N \rightarrow \infty$,

$$E(SD^2\hat{\lambda}) \sim \sum_{i=1}^m w_i \sigma^2(\hat{\lambda}_i) \quad (N \rightarrow \infty). \quad (3.39)$$

In order to demonstrate that for large sample sizes in all age groups $E \hat{\sigma}_{R,\lambda}^2$ is approximately equal to σ_λ^2 , the unconditional variance of $\hat{\lambda}$ is split up into conditional components. Using $\text{Var } \hat{\lambda} = E\hat{\lambda}^2 - (E\hat{\lambda})^2$ and $E\hat{\lambda}^2 = \sum_{i=1}^m w_i E(\hat{\lambda}_i^2)$,

where $E(\hat{\lambda}_i^2) = \text{Var } \hat{\lambda}_i + (E\hat{\lambda}_i)^2$, it is readily seen that

$$\text{Var } \hat{\lambda} = \sum_{i=1}^m w_i \text{Var } \hat{\lambda}_i + \sum_{i=1}^m w_i (E\hat{\lambda}_i)^2 - (E\hat{\lambda})^2.$$

Since $E\hat{\lambda}_i \rightarrow \lambda_i$ and $E\hat{\lambda} \rightarrow \lambda$ as $N \rightarrow \infty$ (see (3.38)) it follows that

$$\{\text{Var } \hat{\lambda} - \sum_{i=1}^m w_i \text{Var } \hat{\lambda}_i\} \rightarrow \sum_{i=1}^m w_i \lambda_i^2 - \lambda^2 \quad (N \rightarrow \infty).$$

The spatial variance $\sigma_\lambda^2 = \sum_{i=1}^m w_i (\lambda_i - \lambda)^2$ and thus it follows that

$$\{\text{Var } \hat{\lambda} - \sum_{i=1}^m w_i \text{Var } \hat{\lambda}_i\} \rightarrow \sigma_\lambda^2 \quad (N \rightarrow \infty). \quad (3.40)$$

So far only some properties of the non-repeated two-stage sampling procedure ($n=1$) have been established. The asymptotic properties (3.38), (3.39) and (3.40) are further used to assess the expected values of the estimators $\hat{\lambda}_R$ and $\hat{\beta}_{R,\lambda}^2$ in the random design. The random design provides n independent realisations of the random vector $(I, \lambda, \hat{\lambda}, \text{SD}^2\hat{\lambda})$. These realisations and associated random variables are written as¹⁾ $(i_j, \lambda_{i_j}, \hat{\lambda}_{i_j}, \text{SD}^2\lambda_{i_j})$ ($j=1, \dots, n$). The $\hat{\lambda}_{i_j}$ ($j=1, \dots, n$) are independent random variables; the distribution of $\hat{\lambda}_{i_j}$ ($j=1, \dots, n$) equals the distribution of $\hat{\lambda}$. Therefore it follows immediately that $E[\frac{1}{n} \sum_{j=1}^n \hat{\lambda}_{i_j}] = E\hat{\lambda}$. Hence by definition of $\hat{\lambda}_R$ and using (3.38) it follows immediately that

$$E(\hat{\lambda}_R) \rightarrow \lambda \quad (N \rightarrow \infty). \quad (3.41)$$

In an analogous way it is readily seen from (3.39) that

$$E\left(\frac{1}{n} \sum_{j=1}^n \text{SD}^2\hat{\lambda}_{i_j}\right) \sim \sum_{i=1}^m w_i \sigma^2(\hat{\lambda}_i) \quad (N \rightarrow \infty). \quad (3.42)$$

¹⁾ The λ_{i_j} , $\hat{\lambda}_{i_j}$ should not be confused with the λ_i and $\hat{\lambda}_i$ in the fixed design: the composed indices refer to the random design.

The sample variance $\frac{1}{n-1} \sum_{j=1}^n (\hat{\lambda}_{1,j} - \hat{\lambda}_R)^2$ is an unbiased estimator of $\text{Var } \hat{\lambda}$, and thus it follows from (3.40) that

$$\left\{ E \left(\frac{1}{n-1} \sum_{j=1}^n (\hat{\lambda}_{1,j} - \hat{\lambda}_R)^2 \right) - \frac{1}{n} \sum_{i=1}^m w_i \text{Var } \hat{\lambda}_{1,i} \right\} \rightarrow \sigma_\lambda^2 \quad (n \rightarrow \infty). \quad (3.43)$$

For $N \rightarrow \infty$ it is plausible that $\sum_{i=1}^m w_i \sigma^2(\hat{\lambda}_{1,i})$ is approximately equal to $\sum_{i=1}^m w_i \text{Var } \hat{\lambda}_{1,i}$ and therefore it follows from (3.42) and (3.43) that for large sample sizes in all age groups the expected value of

$$\frac{1}{n-1} \sum_{j=1}^n (\hat{\lambda}_{1,j} - \hat{\lambda}_R)^2 - \frac{1}{n} \sum_{j=1}^n \text{SD}^2 \hat{\lambda}_{1,j}$$

is close to the parameter value σ_λ^2 . For the present need the use of the estimators $\hat{\lambda}_R$ and $\hat{\sigma}_{R,\lambda}^2$ is sufficiently motivated.

The variation of the spline approximations $\hat{\lambda}_{1,j}$ ($j=1, \dots, n$) in the random design is the result of the influence of three factors: the spatial variation in the trend of the transmission intensity as described by σ_λ^2 , the statistical fluctuations due to the random sampling of n spline approximations $\lambda_{1,j}$ ($j=1, \dots, n$) and the statistical errors in the estimates $\hat{\lambda}_{1,j}$ due to the sampling of individuals. To study that part of the variation in $\hat{\lambda}_{1,j}$ ($j=1, \dots, n$) that can be explained by the sampling of subpopulations and the sampling of individuals in these subpopulations concepts are used similar to the concepts defined in the previous section. In a random sample design we define (see also (3.34)):

$$V_{R,tot} = \text{total variance} = \frac{1}{n-1} \sum_{j=1}^n (\hat{\lambda}_{1,j} - \hat{\lambda}_R)^2 \quad (3.44)$$

$$V_{R,spat} = \text{spatial variance} = \hat{\sigma}_{R,\lambda}^2 \quad (3.45)$$

$$V_{R,err} = \text{error variance} = \frac{1}{n} \sum_{j=1}^n \text{SD}^2 \hat{\lambda}_{1,j}. \quad (3.46)$$

Using this convention the total variance $V_{R,tot}$ can be written as

$$V_{R,tot} = V_{R,spat} + V_{R,err}. \quad (3.47)$$

The proportion explained variance ¹⁾ is defined by

$$E_R = \frac{V_{R,err}}{V_{R,tot}}. \quad (3.48)$$

Estimators of Γ and σ^2_γ , written as $\hat{\Gamma}_R$ and $\hat{\sigma}^2_{R,\gamma}$ are completely similar in formula as the estimators of Λ and σ^2_λ i.e. in (3.32) and (3.34) the $\hat{\lambda}_{1,j}$ has to be substituted by $\hat{\gamma}_{1,j}$ ($j=1, \dots, n$). The variance of $\hat{\Gamma}_R$ is estimated by (see (3.33))

$$SD^2\hat{\Gamma}_R = \frac{1}{n(n-1)} \sum_{j=1}^n (\hat{\gamma}_{1,j} - \hat{\Gamma}_R)^2. \quad (3.49)$$

Furthermore the total variance, the spatial variance, the error variance and the proportion explained variance of $\hat{\gamma}_{1,j}$ ($j=1, \dots, n$) are defined by formulae completely analogous to the corresponding variance concepts with regard to $\hat{\lambda}_{1,j}$ ($j=1, \dots, n$).

Trend-change test for a non-homogeneous population (random design)

To test in a time point t years before survey date the hypothesis

$H_0: \Gamma = 0$, against the two-sided alternative $H_1: \Gamma \neq 0$, the statistic $T_R(t)$ is used

$$T_R(t) = \frac{\hat{\Gamma}_R}{SD\hat{\Gamma}_R}. \quad (3.50)$$

For large sample sizes in the age groups and n sufficiently large $T_R(t)$ is

¹⁾ Note that the proportion explained variance E_R may assume a value > 1 .

treated as an observed value of a standardized normal variable. For the detection of periods of change in the average (spline) trend of the transmission intensity the same procedure is followed as described for the fixed design. The p-value function associated with the random design is written as $P_R(t)$.

3.3.3 Relationship between the analyses based on an F-design and on an R-design

If cross-sectional surveys are carried out in n homogeneous subpopulations Π_1, \dots, Π_n then an analysis based on an F-design as well as on an R-design might be performed. When the analysis is based on the theory of an F-design then the conclusions of the analysis are restricted to the combined population $\Pi_F = \bigcup_{j=1}^n \Pi_j$. If it is acceptable to assume that the subpopulations are a random sample (with replacement) from m homogeneous subpopulations, then an analysis based on an R-design is justified. The population which consists of the m homogeneous subpopulations is denoted by Π_R . In most practical situations it is not likely that the sampling of subpopulations has been carried out with specific weighting factors. In such a situation the conclusions of an analysis based on an R-design might concern the population Π_R with equal weightage for the subpopulations.

In the following equal weightage for the subpopulations is assumed in both designs. In case of an F-design the trend function

$$\Lambda_F = \frac{1}{n} \sum_{j=1}^n \lambda_j \quad (3.51)$$

in population Π_F is studied. If the analysis is performed in an R-design, then the statistical inference concerns the trend function in population Π_R ,

$$\Lambda_R = \frac{1}{m} \sum_{i=1}^m \lambda_i. \quad (3.52)$$

In order to detect periods of change in Λ_F and Λ_R the p-value functions $P_F(t)$ and $P_R(t)$ were used. The proportion explained variance of $\hat{\gamma}_j$ in the F-design and the R-design was written as $E_F(t)$ and $E_R(t)$ respectively. The next theorem shows that there exists a simple relationship between the proportion explained variance of $\hat{\gamma}_j$ and these two p-value functions.

Theorem 3.2 Consider an analysis based on an F-design and on an R-design with equal weightage for the subpopulations in each design. The rates of change in Λ_F and Λ_R are denoted by Γ_F and Γ_R ,

$$\Gamma_F = \frac{1}{n} \sum_{j=1}^n \gamma_j \text{ and } \Gamma_R = \frac{1}{m} \sum_{i=1}^m \gamma_i .$$

If the statistics $T_F(t)$ and $T_R(t)$ are used for testing $\Gamma_F = 0$ and $\Gamma_R = 0$ respectively (see (3.31) and (3.50)), then the following two relations indicate that there exists a straight forward relationship between the p-value functions of these two tests and the proportion explained variance of $\hat{\gamma}_j$ in the two designs. Firstly:

$$E_F(t) = E_R(t). \quad (3.53)$$

In words: the proportion explained variance is equal for both designs.

This proportion is written as $E(t)$, i.e. $E(t) = E_F(t) = E_R(t)$. Secondly we have

$$\frac{T_R(t)}{T_F(t)} = \sqrt{E(t)}. \quad (3.54)$$

As a consequence of (3.54) it follows immediately that the following

(in) equalities hold: $E(t) \leq 1 \iff P_R(t) \geq P_F(t).$ (3.55)

Proof In a *fixed design* with equal weightage for the subpopulations we have (cf. (3.15) and (3.17))

$$\hat{\tau}_F = \frac{1}{n} \sum_{j=1}^n \hat{\gamma}_j \text{ and } SD^2 \hat{\tau}_F = \frac{1}{n^2} \sum_{j=1}^n SD^2 \hat{\gamma}_j.$$

The total variance and the error variance of $\hat{\gamma}_j$ ($j=1, \dots, n$) are (see 3.25 and (3.27))

$$V_{F,tot} = \frac{1}{n} \sum_{j=1}^n (\hat{\gamma}_j - \hat{\tau}_F)^2 \text{ and } V_{F,err} = \frac{n-1}{n^2} \sum_{j=1}^n SD^2 \hat{\gamma}_j.$$

In a *random design* the corresponding formulae are

$$\hat{\tau}_R = \frac{1}{n} \sum_{j=1}^n \hat{\gamma}_j \text{ and } SD^2 \hat{\tau}_R = \frac{1}{n(n-1)} \sum_{j=1}^n (\hat{\gamma}_j - \hat{\tau}_R)^2.$$

Thus the statistics $\hat{\tau}_F$ and $\hat{\tau}_R$ are identical. Furthermore it is readily seen from (3.44) and (3.46) that

$$V_{R,tot} = \frac{n}{n-1} V_{F,tot} \text{ and } V_{R,err} = \frac{n}{n-1} V_{F,err}.$$

It follows immediately that $E_F(t) = E_R(t)$, and thus the proportion explained variance of $\hat{\gamma}_j$ ($j=1, \dots, n$) is equal for both designs ($E(t)$).

For the test statistics in the fixed design and the random design we have

$T_F(t) = \hat{\tau}_F / SD \hat{\tau}_F$ and $T_R(t) = \hat{\tau}_R / SD \hat{\tau}_R$. From the equality of $\hat{\tau}_F$ and $\hat{\tau}_R$ and the formulae of $SD \hat{\tau}_F$ and $SD \hat{\tau}_R$ it follows that

$$\frac{T_R(t)}{T_F(t)} = \frac{SD \hat{\tau}_F}{SD \hat{\tau}_R} = \sqrt{\frac{V_{F,err}/(n-1)}{V_{F,tot}/(n-1)}} = \sqrt{E(t)}.$$

The relationship between the proportion explained variance $E(t)$ and the two p-value functions $P_R(t)$ and $P_F(t)$ follows immediately from the above expression. This completes the proof of theorem 3.2.

APPENDIX 4

CORRECTION FOR FADING OF ANTIBODIES AND SENSITIVITY AND SPECIFICITY OF THE SEROLOGICAL TEST

4.1 INTRODUCTION

Disregarding the factor of fading of antibodies, the sensitivity and the specificity of the serological test in the estimation of the trend of the transmission intensity $\lambda_s(t)$ and its rate of change $\gamma_s(t)$, implies that the estimates of these functions are erroneously based on the seropositive age related fractions. In order to account for this bias a clear distinction is made between the seropositivity rate, written as $p^a(A)$, and the cumulative inoculation rate $p(A)$. The biased transmission function based on $p^a(A)$ instead of $p(A)$ is called *antibody related transmission function* and is denoted by $\lambda^a(t)$, the rate of change in $\lambda^a(t)$ is notated as $\gamma^a(t)$. The spline approximations of these biased functions are denoted by $\lambda_s^a(t)$ and $\gamma_s^a(t)$.

The epidemiologic interpretation of the function $\lambda^a(t)$ and its rate of change $\gamma^a(t)$ depends on the relation between " $\lambda^a(t)$ " and " $\lambda(t)$ " and " $\gamma^a(t)$ " and "

$\gamma(t)$ ". These relations depend on the past-sensitivity and the past-specificity of the serological test. The definition of these concepts and their relationship with fading of antibodies, sensitivity and specificity of the serological test is presented in section 4.2.

The interpretation of the antibody related transmission function in section 4.3 relies on two additional assumptions which extend the series stated in Appendix 2. These two assumptions are: firstly the seropositivity rate $p^a(A)$ is increasing with age A and secondly the past-specificity of the serological test is 1. A method is proposed for the interpretation of the course of the antibody related transmission function $\lambda^a(t)$. This method takes into account the unknown value of the past-sensitivity and its unknown rate of change. This correction method is therefore called maximum linear correction (MLC) method for the factor of past-sensitivity.

Generalizations and extensions of the MLC method are presented in section 4.4

In order to avoid ambiguity in the notation and the epidemiological interpretation of mathematical symbols reference is made to Appendix 1, section 1.4; in particular emphasis is placed on the conventions regarding the description of time and age.

4.2 PAST-SENSITIVITY AND PAST-SPECIFICITY AND THEIR RELATIONSHIP WITH SENSITIVITY, SPECIFICITY AND FADING OF ANTIBODIES

For an individual randomly selected from the (sub) population under study the following six events are considered:

m = the individual has experienced at least one infection with micro-organism m in his life¹⁾

¹⁾ In this study we equate an infection with micro-organism m with malaria.

a_m^c = antibodies specific for micro-organism m are

present at the moment of blood collection

s^+ = serological test result is positive

The symbols m^c , a_m^c and s^- denote the complements of m , a_m and s^+ .

The past-sensitivity (η), past-specificity (ξ), sensitivity (η^a) and specificity (ξ^a) of the serological test are defined by the following conditional probabilities (proportions):

$$\begin{aligned} \text{past-sensitivity } \eta &= p[s^+ | m] & \text{past-specificity } \xi &= p[s^- | m^c] \\ \text{sensitivity } \eta^a &= p[s^+ | a_m] & \text{specificity } \xi^a &= p[s^- | a_m^c]. \end{aligned} \quad (4.1)$$

The degree of fading of antibodies is defined by

$$\text{fading of antibodies } f = p[a_m^c | m]. \quad (4.2)$$

Theorem 4.1 The condition $p[s^+ | m-a_m] = p[s^+ | m^c]$ is necessary and sufficient for each of the following two relations to be true (cf. chapter 4, diagram 4.1).

$$\xi = \xi^a \quad (4.3)$$

$$\eta = (1-f)\eta^a + f(1-\xi^a) \quad (4.4)$$

Furthermore

$$p[s^+ | m-a_m] \geq p[s^+ | m^c] \iff \xi \geq \xi^a \iff$$

$$\eta \geq (1-f)\eta^a + f(1-\xi^a) \iff \eta \geq (1-f)\eta^a + f(1-\xi).$$

Proof: The proof of this theorem is given in four parts a), b), c) and d).

a) First an expression for $\xi - \xi^a$ is derived. We have $\xi - \xi^a = (1-\xi^a) - (1-\xi)$.

Substitution of $(1-\xi^a)$ and $(1-\xi)$ in this expression by:

$$\begin{aligned}
1 - \xi^a &= p[s^+ | a_m^c] = \frac{p[s^+ \cap a_m^c]}{p[a_m^c]} = \frac{p[s^+ \cap (m - a_m)] + p[s^+ \cap m^c]}{p[a_m^c]} = \\
&= \frac{p[m - a_m]}{p[a_m^c]} p[s^+ | m - a_m] + \frac{p[m^c]}{p[a_m^c]} p[s^+ | m^c], \text{ and} \tag{4.5}
\end{aligned}$$

$$1 - \xi = p[s^+ | m^c], \text{ results in} \tag{4.6}$$

$$\xi - \xi^a = \frac{p[m - a_m]}{p[a_m^c]} p[s^+ | m - a_m] + \frac{p[m^c] - p[a_m^c]}{p[a_m^c]} p[s^+ | m^c].$$

Using the equality $p[m - a_m] = p[a_m^c] - p[m^c]$ it is readily seen that

$$\xi - \xi^a = \frac{p[m - a_m]}{p[a_m^c]} (p[s^+ | m - a_m] - p[s^+ | m^c]). \tag{4.7}$$

From formula (4.7) it follows immediately that

$$p[s^+ | m - a_m] \geq p[s^+ | m^c] \iff \xi \geq \xi_a.$$

b) The past-sensitivity η is equal to

$$\begin{aligned}
\eta &= p[s^+ | m] = \frac{p[s^+ \cap m]}{p[m]} = \frac{p[s^+ \cap a_m] + p[s^+ \cap (m - a_m)]}{p[m]} = \\
&= \frac{p[a_m]}{p[m]} p[s^+ | a_m] + \frac{p[m - a_m]}{p[m]} p[s^+ | m - a_m]. \tag{4.8}
\end{aligned}$$

Furthermore by definition the following relations hold:

$$f = p[a_m^c | m] = \frac{p[m - a_m]}{p[m]}, \quad 1 - f = p[a_m | m] = \frac{p[a_m]}{p[m]} \text{ and } \eta^a = p[s^+ | a_m].$$

Substitution of f , $1 - f$ and η^a in (4.8) using these expressions results in the general formula

$$\eta = (1-f) \eta^a + f p[s^+ | m-a_m]. \quad (4.9)$$

Formula (4.9) is used in part c) and d) for the completion of the proof.

c) $\eta - (1-f)\eta^a - f(1-\xi) = f p[s^+ | m-a_m] - f(1-\xi)$. Using (4.6) it follows that

$$\eta - (1-f)\eta^a - f(1-\xi) = f (p[s^+ | m-a_m] - p[s^+ | m^c]) \quad (4.10)$$

Hence $p[s^+ | m-a_m] \geq p[s^+ | m^c] \iff \eta \geq (1-f)\eta^a + f(1-\xi)$.

d) $\eta - (1-f)\eta^a - f(1-\xi^a) = f p[s^+ | m-a_m] - f(1-\xi^a)$. Substitution of $1-\xi^a$ by (4.5) results in

$$\eta - (1-f)\eta^a - f(1-\xi^a) = f(p[s^+ | m-a_m] - \frac{p[m-a_m]}{p[a_m^c]} p[s^+ | m-a_m] - \frac{p[m^c]}{p[a_m^c]} p[s^+ | m^c]).$$

Using the equality $p[a_m^c] - p[m-a_m] = p[m^c]$ it is readily seen that

$$\eta - (1-f)\eta^a - f(1-\xi^a) = f \frac{p[m^c]}{p[a_m^c]} (p[s^+ | m-a_m] - p[s^+ | m^c]). \quad (4.11)$$

From (4.11) it follows immediately that

$$p[s^+ | m-a_m] \geq p[s^+ | m^c] \iff \eta \geq (1-f)\eta^a + f(1-\xi^a).$$

This completes the proof of theorem 4.1.

For any serological test which is used for the detection of antibodies a_m we have $p[s^+ | a_m] > p[s^+ | a_m^c]$ i.e. the following relation between the sensitivity η^a and the specificity ξ^a holds

$$\eta^a > 1-\xi^a. \quad (4.12)$$

If it can be assumed that $p[s^+ | m-a_m] \geq p[s^+ | m^c]$ then an analogous relation exists between the past-sensitivity η and the past specificity ξ of the

serological test. From (4.9) it follows that $\eta \geq (1-f)\eta^a + f p[s^+ | m^c]$ or
 $\eta \geq (1-f)\eta^a + f(1-\xi)$.

As $\eta^a > 1-\xi^a$ we thus have $\eta > (1-f)(1-\xi^a) + f(1-\xi)$ (f ≠ 1).

From $p[s^+ | m-a_m] \geq p[s^+ | m^c]$ it follows that (see theorem 4.1) $\xi \geq \xi^a$ and thus $\eta > (1-f)(1-\xi) + f(1-\xi)$. Hence the following relation holds

$$\eta > 1-\xi \quad (f \neq 1). \quad (4.13)$$

For any serological test which is used for the detection of antibodies a_m we can presuppose $p[s^+ | a_m] > p[s^+ | m-a_m]$. Using this relation and equation (4.9) it is easily seen that the past-sensitivity η is smaller than the sensitivity η^a . From $\eta < (1-f)\eta^a + f p[s^+ | a_m]$ and $\eta^a = p[s^+ | a_m]$ it follows that

$$\eta < \eta^a \quad (f \neq 0). \quad (4.14)$$

4.3 INTERPRETATION OF THE ANTIBODY RELATED TRANSMISSION FUNCTION $\lambda^a(t)$

4.3.1 Relationship between $\lambda(t)$ and $\lambda^a(t)$

The objective of this section is firstly to relate the value of the antibody related transmission function $\lambda^a(t)$ to the value of the transmission function $\lambda(t)$ in a time point A years before survey date and secondly to relate the rate of change of $\lambda^a(t)$ to the rate of change of $\lambda(t)$ in that time point.

Theorem 4.2 If the past-specificity $\xi(A)$ of the serological test is 1 then the following relation holds

$$\lambda(A) = \left[\frac{1-\eta(A)}{\eta(A)-p^a(A)} + 1 \right] \left[\lambda^a(A) - \frac{1}{\eta(A)} \frac{d\eta}{dA} \frac{p^a(A)}{1-p^a(A)} \right]. \quad (4.15)$$

Proof: The value of the transmission function $\lambda(t)$ in a time point A years before survey date is (see Appendix 2, (2.11))

$$\lambda(A) = \frac{d}{dA} \left[-e \log(1-p(A)) \right]. \quad (4.16)$$

The antibody related transmission function $\lambda^a(t)$ is the transmission function erroneously based on the seropositivity rate $p^a(A)$ instead of the cumulative inoculation rate $p(A)$, hence $\lambda^a(A)$ is defined by

$$\lambda^a(A) = \frac{d}{dA} \left[-e \log(1-p^a(A)) \right]. \quad (4.17)$$

The assumption of an increasing seropositivity rate $p^a(A)$, see the introduction 4.1, ensures that the antibody related transmission function $\lambda^a(t)$ assumes positive values. The interpretation of the value of $\lambda^a(t)$ depends on the relation between the seropositivity rate $p^a(A)$ and the cumulative inoculation rate $p(A)$. From the general relation

$$p_A[s^+] = p_A[s^+|m] p_A[m] + p_A[s^+|m^c] p_A[m^c],$$

it follows at once that

$$p^a(A) = n(A) p(A) + [1-\xi(A)] [1-p(A)]. \quad (4.18)$$

Because $\xi(A)=1$ and $p(A) < 1$ we have

$$p^a(A) = n(A) p(A) \text{ and } p^a(A) < n(A) \leq 1, \quad (4.19)$$

where $n(A)$ is the age specific past-sensitivity of the serological test.

From (4.16) and (4.17) it follows that

$$\lambda(A) = \frac{\frac{dp}{dA}}{1-p(A)} \text{ and } \lambda^a(A) = \frac{\frac{dp^a}{dA}}{1-p^a(A)}. \quad (4.20)$$

Using (4.19), the rate of change of the seropositivity rate is equal to $\frac{dp^a}{dA} = \eta(A)\frac{dp}{dA} + p(A)\frac{dn}{dA}$. Solving this equation for $\frac{dp}{dA}$ and using the relation $\frac{dp}{dA} = \gamma^a(A)(1-p^a(A))$, see (4.20), it follows that

$$\frac{dp}{dA} = \frac{1}{\eta(A)} \left[\lambda^a(A)(1-p^a(A)) - \frac{dn}{dA} p(A) \right].$$

Substitution of $\frac{dp}{dA}$ in the formula for $\lambda(A)$ and using (4.19) it is readily derived that $\lambda(A)$ is equal to

$$\lambda(A) = \frac{1-p^a(A)}{\eta(A) - p^a(A)} \left[\lambda^a(A) - \frac{\frac{dn}{dA} p^a(A)}{\eta(A)(1-p^a(A))} \right] \quad (4.21)$$

From this equation immediately follows formula (4.15). This completes the proof of theorem 4.2.

Theorem 4.3 If the past-specificity $\xi(A)$ of the serological test is 1 then the following relation holds

$$\gamma(A) = \left[\frac{1-\eta(A)}{\eta(A)-p^a(A)} + 1 \right] \left[\gamma^a(A) + \Delta(A) \right], \quad (4.22)$$

where $\Delta(A)$ is a function defined by

$$\Delta(A) = \Delta_1(A) + \Delta_2(A) + \Delta_3(A), \quad (4.23)$$

with $\Delta_1(A) = \frac{1-\eta(A)}{\eta(A)-p^a(A)} (\lambda^a(A))^2$,

$$\Delta_2(A) = - \frac{2\lambda^a(A)}{\eta(A)-p^a(A)} \frac{dn}{dA} + \frac{1}{\eta(A)^2} \frac{2\eta(A)-p^a(A)}{\eta(A)-p^a(A)} \frac{p^a(A)}{1-p^a(A)} \left(\frac{dn}{dA} \right)^2,$$

$$\Delta_3(A) = - \frac{1}{\eta(A)} \frac{p^a(A)}{1-p^a(A)} \frac{d^2\eta}{dA^2}.$$

Comment upon the term $\Delta(A)$: The direction of the rate of change of the transmission function $\lambda(t)$ in a time point A years before survey date is determined by the sign of $(\gamma^a(A) + \Delta(A))$. Therefore the term $\Delta(A)$

is called the sign disturbance coefficient. This function will be studied further in the next section.

Proof of theorem 4.3: The functions $\gamma(t)$ and $\gamma^a(t)$ are defined by

$$\gamma(t) = \frac{d}{dt} \lambda(t) \quad \gamma^a(t) = \frac{d}{dt} \lambda^a(t). \quad (4.24)$$

From (4.20) it follows that the value of $\gamma(t)$ and $\gamma^a(t)$ in a time point A years before survey date is

$$\gamma(A) = \frac{\frac{d^2 p}{dA^2}}{1-p(A)} + \lambda^2(A) \quad \gamma^a(A) = \frac{\frac{d^2 p^a}{dA^2}}{1-p^a(A)} + [\lambda^a(A)]^2. \quad (4.25)$$

Using (4.19), the second derivative of the seropositivity rate is

$\frac{d^2 p^a}{dA^2} = n(A) \frac{d^2 p}{dA^2} + 2 \frac{dn}{dA} \frac{dp}{dA} + p(A) \frac{d^2 n}{dA^2}$. Solving this equation for $\frac{d^2 p}{dA^2}$ and using

the relations $\frac{d^2 p^a}{dA^2} = [\gamma^a(A) - [\lambda^a(A)]^2](1-p^a(A))$ and $\frac{dp}{dA} = \lambda(A)(1-p(A))$,

we have $\frac{d^2 p}{dA^2} = \frac{1}{n(A)} \left[[\gamma^a(A) - \lambda^a(A)^2](1-p^a(A)) - 2 \frac{dn}{dA}(1-p(A))\lambda(A) - \frac{d^2 n}{dA^2} p(A) \right]$.

Substitution of $\frac{d^2 p}{dA^2}$ in the formula for $\gamma(A)$, see (4.25), and using (4.19) it is readily verified that $\gamma(A)$ is equal to

$$\gamma(A) = \frac{1-p^a(A)}{n(A)-p^a(A)} \left[\gamma^a(A) - \lambda^a(A)^2 - \frac{2}{n(A)} \frac{dn}{dA} \frac{n(A)-p^a(A)}{1-p^a(A)} \lambda(A) - \frac{1}{n(A)} \frac{d^2 n}{dA^2} \frac{p^a(A)}{1-p^a(A)} \right] + \lambda^2(A).$$

Substitution of $\lambda(A)$ in this equation by the formula of $\lambda(A)$ presented in theorem 4.2 results in formula (4.22). This completes the proof of theorem 4.3.

4.3.2 The sign disturbance coefficient $\Delta(A)$

The sign disturbance coefficient $\Delta(A)$ depends on the following related variables (see theorem 4.3, formula (4.23))

$$\Delta(A) = \Delta(p^a(A), \lambda^a(A), \eta(A), \frac{dn}{dA}, \frac{d^2n}{dA^2}).$$

The values $p^a(A)$ and $\lambda^a(A)$ can be estimated from the data. The value $\eta(A)$, its rate of change and its curvature, however, are virtually unknown. Therefore it is impossible to assess the value of $\Delta(A)$ taking into account all five variables. When it is not acceptable to make any assumptions with respect to $(\eta(A), \frac{dn}{dA}, \frac{d^2n}{dA^2})$ then correction for the factor past-sensitivity is impossible and consequently the serological profile of the community $p^a(A)$ cannot give information on the course of the transmission intensity in that community. In this section the range of values the sign disturbance coefficient $\Delta(A)$ may attain in an arbitrary but fixed time point A years before survey date is assessed while disregarding the curvature of the past-sensitivity; i.e. it is assumed that $\frac{d^2n}{dA^2} = 0$ for that particular time point A. Thus in the following only $p^a(A)$, $\lambda^a(A)$ and the value of the past-sensitivity $\eta(A)$ and its rate of change $\frac{dn}{dA}$ are taken into account.

In order to simplify the notation the following abbreviations are used in the sequel (A is arbitrary but fixed):

$$\Delta = \Delta(A), p^a = p^a(A), \lambda^a = \lambda^a(A), \eta = \eta(A).$$

Lemma 4.1 If it can be assumed that

- the past-specificity $\xi(A)$ of the serological test is 1 and
- the sero positivity rate $p^a(A)$ is increasing with age and
- the curvature $\frac{d^2n}{dA^2}$ of the past sensitivity can be disregarded, then it follows that:

(1) If $\eta(A)$ is not changing for age A, i.e. $\frac{dn}{dA} = 0$, then

$$\Delta = \Delta_0 = \frac{1-\eta}{\gamma-p} a [\lambda^a]^2 \quad (4.26)$$

(2) If $\eta(A)$ is decreasing for age A , i.e. $\frac{d\eta}{dA} < 0$, then $\Delta > \Delta_0$ (4.27)

(3) If $\eta(A)$ is increasing for age A , i.e. $\frac{d\eta}{dA} > 0$, then $\Delta_{\min} \leq \Delta < \Delta_0$, (4.28)

$$\text{where } \Delta_{\min} \text{ is defined by } \Delta_{\min} = -\frac{\eta + np^a - p^a}{2np^a} \frac{[\lambda^a]^2}{p^a}. \quad (4.29)$$

Furthermore in case that the rate of change of $\eta(A)$ and its direction are unknown, it follows from (1), (2) and (3) that

$$\Delta \geq \Delta_{\min}. \quad (4.30)$$

Proof: From theorem 4.3 formula (4.23) it follows that $\Delta(A) = \Delta_1(A) + \Delta_2(A)$.

In order to establish the range of values $\Delta(A)$ may attain, it is convenient to consider $\Delta(A)$ as a function of the relative rate of change in $\eta(A)$, i.e. as a function of $r(A)$, where $r(A) = \frac{1}{\eta(A)} \frac{d\eta}{dA}$. As already indicated the symbol A is often suppressed to simplify the notation, thus $r(A)$ is written as r .

The sign disturbance coefficient Δ is easily expressed in r

$$\Delta = ar^2 + br + c, \quad (4.31)$$

$$\text{where } a = \frac{2np^a}{\eta-p^a} \frac{p^a}{1-p^a}, \quad b = -\frac{2\eta}{\eta-p^a} \lambda^a, \quad c = \frac{1-\eta}{\eta-p^a} [\lambda^a]^2.$$

As a function of r , Δ is a polynomial of degree 2. The coefficient a in (4.31) is always larger than zero. As $\lambda(A)$ is strictly positive it follows from theorem 4.2 that r may assume values in the range $(-\infty, \frac{1-p^a}{p^a} \lambda^a)$, i.e.

$$r \leq \frac{1-p^a}{p^a} \lambda^a. \quad (4.32)$$

The sign disturbance coefficient Δ as a function of r takes on its minimum value if $r=r_{\min}$, with r_{\min} given by

$$r_{\min} = \frac{\eta}{2np^a} \frac{1-p^a}{p^a} \lambda^a. \quad (4.33)$$

It has been assumed that the seropositivity rate $p^a(A)$ increases with age.

Therefore we have $\lambda^a(A) > 0$ and thus $r_{\min} > 0$. Comparison of (4.32) with (4.33) shows that r may attain the value r_{\min} and furthermore $r < 2r_{\min}$.

For $r = 0$ and $r = r_{\min}$, Δ assumes the value Δ_0 and Δ_{\min} , where $\Delta_0 = \frac{1-\eta}{n-p^a} [\lambda^a]^2$ and $\Delta_{\min} = -\frac{n+np^a-p^a}{2n-p^a} \frac{[\lambda^a]^2}{p^a}$.

From the shape of the parabola ($a > 0$) it follows at once that:

$$\text{If } r < 0 \text{ then } \Delta > \Delta_0$$

$$\text{If } r = 0 \text{ then } \Delta = \Delta_0$$

$$\text{If } r > 0 \text{ then } \Delta_{\min} \leq \Delta < \Delta_0.$$

This completes the proof of lemma 4.1.

4.3.3 The critical past-sensitivity $\eta_c(A)$

In lemma 4.1 bounds Δ_0 and Δ_{\min} have been determined for the value the sign disturbance coefficient Δ may attain. These bounds however only account for the rate of change $\frac{d\eta}{dA}$ of the past-sensitivity and its direction. The Δ_0 and Δ_{\min} still depend on the unknown value η of the past-sensitivity. The next step is to determine the sign of $\gamma^a + \Delta_0$ and the sign of $\gamma^a + \Delta_{\min}$ respectively, while taking into account all possible values the past-sensitivity η may assume. The Δ_0 is strictly positive ($\eta \neq 1$) and Δ_{\min} is strictly negative. Hence there is no problem to determine the sign of $(\gamma^a + \Delta_0)$ for $\gamma^a \geq 0$ and the sign of $(\gamma^a + \Delta_{\min})$ for $\gamma^a \leq 0$. Therefore the functions to be investigated further are $(\gamma^a + \Delta_0)$ for $\gamma^a < 0$ and $(\gamma^a + \Delta_{\min})$ for $\gamma^a > 0$.

Let the functions $f(\eta)$ and $g(\eta)$ be defined by

$$f(\eta) = \gamma^a + \Delta_0 \quad (\gamma^a < 0; p^a < \eta \leq 1) \quad (4.34)$$

$$g(r) = \gamma^a + \Delta_{\min} \quad (\gamma^a > 0; p^a < \eta \leq 1). \quad (4.35)$$

The sign of $f(n)$ and the sign of $g(n)$ depend on the value n of the past-sensitivity. Values of n for which $f(n) = 0$ respectively $g(n) = 0$ are *critical values of the past-sensitivity*. In these values $f(n)$ and $g(n)$ respectively changes sign.

Lemma 4.2 Let $f(n)$ and $g(n)$ be functions defined by (4.34) and (4.35). With respect to the sign of these functions we have:

(1) The function $f(n)$ assumes the value zero in interval $p^a < n \leq 1$. let for $\gamma^a < 0$ the value n_c be defined by $f(n_c) = 0$ or equivalently

$$n_c = \frac{[\lambda^a]^2 - \gamma^a p^a}{[\lambda^a]^2 - \gamma^a} \quad \gamma^a < 0. \quad (4.36)$$

Using the critical value n_c defined by (4.36) it follows that

$$\begin{aligned} \text{if } n < n_c \text{ then } f(n) > 0, \\ \text{if } n > n_c \text{ then } f(n) < 0. \end{aligned} \quad (4.37)$$

(2) The course of the function $g(n)$ is more complicated:

a) The function $g(n)$ is strictly positive $\iff \gamma^a > \frac{[\lambda^a]^2}{p^a(2-p^a)}$ (4.38)

b) The function $g(n)$ is strictly negative $\iff \gamma^a \leq [\lambda^a]^2$ (4.39)

c) For values $\gamma^a > 0$ for which $[\lambda^a]^2 < \gamma^a \leq \frac{[\lambda^a]^2}{p^a(2-p^a)}$, the function (4.40)

$g(n)$ takes on the value zero in interval $p^a < n \leq 1$. let for γ^a in this interval the value n_c be defined by $g(n_c) = 0$ or equivalently

$$n_c = \frac{[\lambda^a]^2 - \gamma^a p^a}{[1+(p^a)^{-1}] [\lambda^a]^2 - 2\gamma^a}, \text{ where } [\lambda^a]^2 < \gamma^a \leq \frac{[\lambda^a]^2}{p^a(2-p^a)}. \quad (4.41)$$

Using the critical value n_c defined by (4.41) it follows that

$$\begin{aligned} \text{If } n < n_c \text{ then } g(n) > 0 \\ \text{If } n > n_c \text{ then } g(n) < 0. \end{aligned} \quad (4.42)$$

Remarks on the critical value η_c . In part (1) of lemma 4.2 the critical value η_c is defined for $\gamma^a < 0$. In part 2 the critical value η_c is defined for $\gamma^a > 0$ in the interval defined by (4.40). The formula for η_c (4.41) is in this situation different from formula (4.36). The critical values η_c defined in lemma 4.2 are called *critical past-sensitivity*; these values play a role in the interpretation of $\gamma^a < 0$ and $\gamma^a > 0$ (next section).

Proof of lemma 4.2:

(1) From the definition of $f(\eta)$ and using the expression for Δ_0 (see lemma 4.1) we have $f(\eta) = \gamma^a + \frac{1-\eta}{n-p^a} [\lambda^a]^2$ ($\gamma^a < 0$; $p^a < \eta \leq 1$).

It is readily verified that the function $f(\eta)$ is monotone decreasing for $p^a < \eta \leq 1$ and that $f(\eta)$ takes on the value zero for $\eta = \eta_c$ with η_c given by (4.36). As $f(\eta)$ is monotone decreasing, relation (4.37) follows immediately.

(2) From the definition of $g(\eta)$ and the formula for Δ_{\min} (see lemma 4.1) it follows that $g(\eta) = \gamma^a - \frac{n+np^a - p^a}{2n - p^a} \frac{[\lambda^a]^2}{p^a}$ ($\gamma^a > 0$; $p^a < \eta \leq 1$).

It is readily verified that the function $g(\eta)$ is monotone decreasing for $p^a < \eta \leq 1$. The values of $g(\eta)$ in the "end-points" of the interval are

$$g(p^a) = \gamma^a - [\lambda^a]^2, \quad g(1) = \gamma^a - \frac{[\lambda^a]^2}{p^a(2-p^a)}.$$

The function $g(\eta)$ is strictly positive $\Leftrightarrow g(1) > 0$ and strictly negative $\Leftrightarrow g(p^a) \leq 0$. The function $g(\eta)$ assumes the value zero in the interval $p^a < \eta \leq 1 \Leftrightarrow g(p^a) > 0$ and $g(1) \leq 0$. Furthermore it is easily verified that $g(\eta_c) = 0$ with η_c defined by (4.41). As $g(\eta)$ is monotone decreasing, relation (4.42) follows immediately.

4.3.4 The sign of $\gamma(A)$

The interpretation of the rate of change of the antibody related transmission function $\lambda^a(t)$ in a time point A years before survey date depends on

the value and course of the past-sensitivity for the corresponding age A. In this section four situations are considered (A fixed), these are:

- The value of the past-sensitivity $\eta(A)$ is unknown, its rate of change is disregarded, i.e. it supposed that $\frac{d\eta}{dA} = 0$ in time point A
- Past-sensitivity $\eta(A)$ decreases with age, its value and the rate of the decrease are unknown
- Past-sensitivity $\eta(A)$ increases with age, its value and the rate of the increase are unknown
- Past-sensitivity $\eta(A)$ changes with age, its value, the rate of change and the direction of change are unknown.

The theorems derived in this section are based on theorem 4.3, lemma 4.1 and lemma 4.2. The assumptions stated in lemma 4.1 are therefore also presupposed in this section. In particular the curvature of the past-sensitivity is disregarded throughout ($\frac{d^2\eta}{dA^2} = 0$). As in section 4.4 and 4.5 the symbol A is often suppressed in the formulae. Furthermore the critical past-sensitivity η_c is considered to be one function although defined by different formulae in different intervals i.e. η_c is defined by

$$\eta_c = \frac{[\lambda^a]^2 - \gamma^a p^a}{[\lambda^a]^2 - \gamma^a} \text{ for } \gamma^a < 0 \text{ and } \eta_c = \frac{[\lambda^a]^2 - \gamma^a p^a}{[1+(p^a)^{-1}] [\lambda^a]^2 - 2\gamma^a} \text{ for } \ell < \gamma^a \leq u,$$

where ℓ and u are defined by

$$\ell = [\lambda^a]^2 \quad u = \frac{[\lambda^a]^2}{p^a(2-p^a)} . \quad (4.44)$$

The ℓ and u are called *lowerbound* and *upperbound*. These bounds are used for the interpretation of a "decreasing" antibody related transmission function ($\gamma^a > 0$) in the situation that $\frac{d\eta}{dA}$ assumes an arbitrary and unknown value. The situation $\frac{d\eta}{dA} = 0$, however, is considered first in the following theorem.

Theorem 4.4 Assuming $\xi(A) = 1$, $\eta(A)$ is unknown and $\frac{d\eta}{dA} = 0$ in a time point A years before survey date, the following relations hold (cf. diagram 4.2 in chapter 4):

- a) If $\gamma^a > 0$ then $\gamma > 0$;
- b) If $\gamma^a = 0$ then $\gamma > 0$ ($\eta \neq 1$);
- c) If $\gamma^a < 0$ then we have:
 - c₁) if $\eta < \eta_c$ then $\gamma > 0$ (opposite sign),
 - c₂) if $\eta > \eta_c$ then $\gamma < 0$ (equal sign).

Proof: From theorem 4.3 and lemma 4.1 it follows that

$$\text{sign } \gamma = \text{sign } (\gamma^a + \Delta), \text{ where } \Delta = \Delta_0 = \frac{1-\eta}{\eta-p} [\lambda^a]^2.$$

If $\gamma^a \geq 0$ then $\gamma^a + \Delta_0 > 0$, because Δ_0 is strictly positive ($\eta \neq 1$), and thus $\gamma > 0$. Furthermore if $\gamma^a < 0$ then $\text{sign } (\gamma^a + \Delta_0) = \text{sign } f(\eta)$ (see formula (4.34))

From lemma 4.2 formula (4.37) it follows immediately that:

if $\eta < \eta_c$ then $\text{sign } (\gamma^a + \Delta_0) > 0$ and thus $\gamma > 0$,

if $\eta > \eta_c$ then $\text{sign } (\gamma^a + \Delta_0) < 0$ and thus $\gamma < 0$.

This completes the proof of theorem 4.4.

The following two lemma's are used to study the situation $\frac{d\eta}{dA} \neq 0$ in a time point A years before survey date.

Lemma 4.3 Assuming $\xi(A) = 1$, $\eta(A)$ is unknown and $\frac{d\eta}{dA} < 0$ in a time point A years before survey date, the following relations hold (cf. diagram 4.3^a in chapter 4):

- a) If $\gamma^a > 0$ then $\gamma > 0$;
- b) If $\gamma^a = 0$ then $\gamma > 0$;
- c) If $\gamma^a < 0$ then we have:
 - c₁) if $\eta < \eta_c$ then $\gamma > 0$ (opposite sign),
 - c₂) if $\eta > \eta_c$ then sign γ is uncertain.

Proof: From theorem 4.3 and lemma 4.1 it follows that $\text{sign } \gamma = \text{sign } (\gamma^a + \Delta)$, where $\Delta > \Delta_0$ and $\Delta_0 = \frac{1-\eta}{\eta-p} [\lambda^a]^2$.

If $\gamma^a \geq 0$ then $\gamma^a + \Delta$ is strictly positive and thus $\gamma > 0$. Furthermore as in theorem 4.4 we have: if $\gamma^a < 0$ then $\text{sign } (\gamma^a + \Delta) = \text{sign } f(\eta)$. Using lemma 4.2 it is readily seen that the following statements hold.

If $\eta < \eta_c$ then $f(\eta) > 0$ and since $\Delta > \Delta_0$ it follows that $\text{sign } (\gamma^a + \Delta) > 0$ and thus $\text{sign } \gamma > 0$. Furthermore if $\eta > \eta_c$ then $f(\eta) < 0$ and thus $(\gamma^a + \Delta) < 0$. However, since $\Delta > \Delta_0$ the sign of $(\gamma^a + \Delta)$, and thereby the sign of γ , is now uncertain. This completes the proof of lemma 4.3.

Lemma 4.4 Assuming $\xi(A) = 1$, $\eta(A)$ is unknown and $\frac{d\eta}{dA} > 0$ in a time point A years before survey date, the following relations hold (cf. diagram 4.3^b in chapter 4):

a) If $\gamma^a > 0$ then it can be derived that:

- a₁) if $\gamma^a > u$ then $\gamma > 0$,
- a₂) if $\gamma^a \leq l$ then sign γ is uncertain,
- a₃) if $l < \gamma^a \leq u$ then we have:
 - if $\eta < \eta_c$ then $\gamma > 0$,
 - if $\eta > \eta_c$ then sign γ is uncertain.

b) If $\gamma^a = 0$ then sign γ is uncertain;

c) If $\gamma^a < 0$ then we have:

- c₁) if $\eta < \eta_c$ then sign γ is uncertain,
- c₂) if $\eta > \eta_c$ then $\gamma < 0$.

Proof: From theorem 4.3 and lemma 4.1 it follows that $\text{sign } \gamma = \text{sign } (\gamma^a + \Delta)$,

where $\Delta_{\min} \leq \Delta < \Delta_0$.

Since Δ_{\min} is strictly negative and Δ_0 is strictly positive it is obvious that $\gamma^a = 0$ implies that sign γ is uncertain. In order to prove a) and c) lemma 4.2 is used.

ad a) If $\gamma^a > 0$ then $\gamma^a + \Delta_{\min} = g(n)$. This function $g(n)$ is strictly positive $\iff \gamma^a > u(A)$. Therefore if $\gamma^a > u(A)$ then $\gamma^a + \Delta_{\min} > 0$ and consequently since $\Delta \geq \Delta_{\min}$ it follows that sign $(\gamma^a + \Delta) > 0$ and thus $\gamma > 0$.

The function $g(n)$ is definite negative $\iff \gamma^a \leq 1(A)$. Therefore if $\gamma^a \leq 1(A)$ then $\gamma^a + \Delta_{\min} < 0$ and since $\Delta \geq \Delta_{\min}$ the sign of $(\gamma^a + \Delta)$ is uncertain.

If $1(A) < \gamma^a \leq u(A)$ then $g(n)$ takes on the value zero. Using (4.42) we have: if $n < n_c$ then $\gamma^a + \Delta_{\min} > 0$ and it follows that sign $(\gamma^a + \Delta) > 0$ and thus $\gamma > 0$. If on the other hand $n > n_c$ then $\gamma^a + \Delta_{\min} < 0$ and this implies that sign $(\gamma^a + \Delta)$, and thus sign γ , is uncertain.

ad c) In case that $\gamma^a < 0$ then $\gamma^a + \Delta_0 = f(n)$. Using (4.37) we have: if $n < n_c$ then $\gamma^a + \Delta_0 > 0$ and since $\Delta < \Delta_0$ it follows that sign $(\gamma^a + \Delta)$ and thereby sign γ is uncertain. Furthermore if $n > n_c$ then $\gamma^a + \Delta_0 < 0$ and it follows that sign $(\gamma^a + \Delta) < 0$ and thus $\gamma < 0$. This completes the proof of lemma 4.4.

Theorem 4.5 Assuming $\xi(A) = 1$, $n(A)$ and $\frac{dn}{dA}$ unknown in a time point A years before survey date, the following relations hold (cf. diagram 4.4 in chapter 4):

a) If $\gamma^a > 0$ then we have:

a₁) if $\gamma^a > u$ then $\gamma > 0$,

a₂) if $\gamma^a \leq l$ then sign γ is uncertain,

a₃) if $l < \gamma^a \leq u$ then we have:

if $n < n_c$ then $\gamma > 0$,

if $n > n_c$ then sign γ is uncertain.

b) If $\gamma^a = 0$ then sign γ is uncertain;

c) If $\gamma^a < 0$ then sign γ is uncertain.

Proof: This theorem follows immediately from lemma 4.3 and lemma 4.4.

The value of the past-sensitivity $\eta(A)$, its rate of change and the direction of the change are virtually unknown. Therefore theorem 4.5 has been used as a guide to develop a procedure for the detection of a change in the trend of transmission in a time point A years before survey date.

Apparently $\gamma^a(A) < 0$ - that is an increasing¹⁾ antibody related transmission function A years before survey date - cannot be interpreted. A negative value $\gamma^a(A)$ can always be explained by the unknown value $\eta(A)$ and its rate of change $\frac{d\eta}{dA}$. However if $\gamma^a(A) > 0$ then we have (see theorem 4.5):

if $(\gamma^a(A) > u(A))$ or $(\lambda(A) < \gamma^a(A) \leq u(A) \text{ and } \eta(A) < \eta_c(A))$ then $\gamma(A) > 0$.

If $\gamma^a(A)$ takes on a value in the interval $(\eta_c(A), u(A))$ then an estimate of the critical past-sensitivity $\eta_c(A)$ might be used in making the decision whether $\gamma(A)$ is larger than zero or whether sign $\gamma(A)$ is uncertain.

Such a decision, however, will always be *subjective* since the actual value of $\eta(A)$ is unknown. Only in the situation where the seropositivity rate $p^a(A)$ is not too near to 1 and $\eta_c(A)$ is relatively large (close to 1) we might decide that the actual unknown past-sensitivity $\eta(A)$ is smaller than the critical past-sensitivity $\eta_c(A)$ and consequently declare $\gamma(A) > 0$. In order to avoid such a subjective decision only the upperbound $u(A)$ is used for the interpretation of a decrease in $\lambda^a(t)$ in a time point A years before survey date, i.e. the basic relation used in the sequel is

$$\text{if } \gamma^a(A) > u(A) \text{ then } \gamma(A) > 0. \quad (4.45)$$

In the applications in chapter 5 a two-step procedure has been used for the detection of a downward trend of the transmission intensity in a time

¹⁾ Note: The course of $\lambda^a(t)$ and $\lambda(t)$ is described from the past to the survey date.

point A years before survey date. Step 2 of this procedure uses relation (4.45) to interprete a decrease in the spline trend $\hat{\lambda}_s^a(t)$. As (4.45) accounts for any linear course of the past-sensitivity $\eta(A)$ with age, the correction method in step 2 of the procedure has been called *maximum linear correction (MLC)* method for the factor of past-sensitivity.

Two-step procedure for the detection of a downward trend of the transmission intensity in a homogeneous population

Step 1: The "trend-change" test introduced in Appendix 2 is used to determine periods before survey date in which the spline approximation $\hat{\lambda}_s^a(t)$ is significantly decreasing (one-sided test $p \leq 0.05$). Time points A in these periods are analysed further in step 2 of the procedure. If $p > 0.05$ for each time point t before survey date ($0 < t < T_k$) then the decision is made that the serological data do not provide an indication for a downward trend of the transmission intensity and step 2 is not performed.

Step 2: If the rate of decrease $\hat{\gamma}_s^a(t)$ is sufficiently large, i.e. larger than an upperbound $\hat{u}(A)$, to be defined below, then it is decided that the serological data provide evidence for a downward trend of the transmission intensity A years before survey date.

An estimate $\hat{u}(A)$ of $u(A)$ is obtained by substituting appropriate estimates of $\lambda^a(A)$ and $p^a(A)$ in the formula for $u(A)$ (see (4.44))

$$\hat{u}(A) = \frac{[\hat{\lambda}_s^a(A)]^2}{\hat{p}^a(A)(2-\hat{p}^a(A))}. \quad (4.46)$$

In the applications the seropositivity rate $p^a(A)$ is simply estimated by linear interpolation using the observed seropositivity rates $\hat{p}^a(A)$ of the two nearest age-groups.

Relation (4.45) is a relation between $\gamma^a(A)$ and $\gamma(A)$. The question arises:

is there an analogous relationship between $\gamma_s^a(A)$ and $\gamma_s(A)$?

The next section is i.a. concerned with this problem.

4.4 EXTENSIONS AND GENERALIZATIONS

4.4.1 Constant past-sensitivity η and constant past-specificity ξ

If the past-specificity $\xi(A)$ of the serological test is 1 and the past-sensitivity $\eta(A)$ is constant, $\eta(A) = \eta$, then the following relations have been derived (see theorema 4.2 and 4.3):

$$\lambda(A) = \left[\frac{1-\eta}{\eta-p^a(A)} + 1 \right] \lambda^a(A) \text{ and} \quad (4.47)$$

$$\gamma(A) = \left[\frac{1-\eta}{\eta-p^a(A)} + 1 \right] [\gamma^a(A) + \Delta(A)], \text{ with } \Delta(A) = \frac{1-\eta}{\eta-p^a(A)} \lambda^a(A)^2. \quad (4.48)$$

It appears that the formulae (4.47) and (4.48) hold irrespective the value of the constant past-specificity ξ . This is expressed by the following theorem.

Theorem 4.6 Assuming a constant past-sensitivity η and a constant past-specificity ξ then the relationship between $\lambda(A)$ and $\lambda^a(A)$ and between $\gamma(A)$ and $\gamma^a(A)$ is given by (4.47) and (4.48) respectively¹⁾. Furthermore theorem 4.4 remains valid ($0 < \eta < 1$, $0 < \xi \leq 1$).

Proof: The general relation between $p^a(A)$ and $p(A)$ is given by (4.18). Using the assumption $\eta(A) = \eta$ and $\xi(A) = \xi$ it follows that

$$p^a(A) = \eta p(A) + (1-\xi)(1-p(A)).$$

¹⁾ Note that the seropositivity rate $p^a(A)$ depends on the past-specificity ξ .

Using this relation it is readily seen that

$$p(A) = \frac{p^a(A) - (1-\xi)}{\eta - (1-\xi)}, \quad \frac{dp}{dA} = \frac{1}{\eta - (1-\xi)} \frac{dp^a}{dA} \quad \text{and} \quad \frac{d^2 p}{dA^2} = \frac{1}{\eta - (1-\xi)} \frac{d^2 p^a}{dA^2}.$$

Substitution of $p(A)$ and $\frac{dp}{dA}$ in the formula for $\lambda(A)$ presented in (4.20), results in

$$\lambda(A) = \frac{\frac{dp}{dA}}{\frac{dA}{\eta - p^a(A)}} = \frac{1-p^a(A)}{\eta - p^a(A)} \frac{\frac{dp}{dA}}{1-p^a(A)}. \quad \text{Using the expression for } \lambda^a(A), \text{ see (4.20),}$$

we thus have $\lambda(A) = \frac{1-p^a(A)}{\eta - p^a(A)} \lambda^a(A)$. Relation (4.47) follows immediately from this last expression.

Substitution of $p(A)$ and $\frac{d^2 p}{dA^2}$ in the formula for $\gamma(A)$ presented in (4.25), results in

$$\gamma(A) = \frac{\frac{d^2 p}{dA^2}}{\frac{dA^2}{\eta - p^a(A)}} + \lambda^2(A). \quad \text{Using the expression for } \lambda(A) \text{ it follows that}$$

$$\gamma(A) = \frac{1-p^a(A)}{\eta - p^a(A)} \frac{\frac{d^2 p}{dA^2}}{1-p^a(A)} + \left[\frac{1-p^a(A)}{\eta - p^a(A)} \right]^2 \lambda^a(A)^2. \quad \text{And thus we have}$$

$$\gamma(A) = \left[\frac{1-p^a(A)}{\eta - p^a(A)} \right] \left[\frac{\frac{d^2 p}{dA^2}}{\frac{dA^2}{1-p^a(A)}} + \lambda^a(A)^2 - \lambda^a(A)^2 + \frac{1-p^a(A)}{\eta - p^a(A)} \lambda^a(A)^2 \right].$$

Using the formula for $\gamma^a(A)$ presented in (4.25), relation (4.48) is readily derived. Since theorem 4.4 is based on formula (4.48), this theorem remains valid in case the past-specificity $\xi(A)$ is constant and not equal to 1. This completes the proof of theorem 4.6.

4.4.2 Variable past-sensitivity $\eta(A)$ and variable past-specificity $\xi(A)$

The main objective is to extend theorem 4.5. As distinct from the assumption in that theorem we neither presuppose here a past-specificity $\xi(A) = 1$ nor a monotone increasing seropositivity rate $p^a(A)$ with age nor $\frac{d^2 \eta}{dA^2} = 0$. In section 4.4.2.1 parts of theorem 4.2, theorem 4.3 and lemma 4.1,

lemma 4.2 are generalized. The results are used to derive theorem 4.7. This theorem is used in the remaining sections for the interpretation of a decrease in $\lambda^a(t)$, $\lambda_s^a(t)$ and $\Lambda_s^a(t)$ respectively.

4.4.2.1. Basic theorem

Lemma 4.5 let $\tau_1(t)$ and $\tau_2(t)$ be functions defined for $0 \leq t \leq T_k$, with continuous derivatives denoted by $\delta_1(t)$ and $\delta_2(t)$. T_k is an arbitrary positive value. Furthermore, let $p_1(A)$ and $p_2(A)$ be functions defined by

$$p_1(A) = \frac{A}{1-k_1} \exp\left(-\int_0^A \tau_1(t) dt\right), \text{ and } p_2(A) = \frac{A}{1-k_2} \exp\left(-\int_0^A \tau_2(t) dt\right),$$

where $0 \leq A \leq T_k$ and k_1 and k_2 are constants, $0 < k_1 \leq 1$, $0 < k_2 \leq 1$.

Let $\kappa(A)$ be the ratio of $p_1(A)$ and $p_2(A)$: $\kappa(A) = \frac{p_1(A)}{p_2(A)}$, $p_2(A) \neq 0$.

For a time point A with $p_2(A) \neq 0$ the following relations hold¹⁾ between " $\tau_1(A)$ and $\tau_2(A)$ " and " $\delta_1(A)$ and $\delta_2(A)$ " (in order to simplify the notation the time point A is omitted):

$$(1) \quad \tau_2 = \left[\frac{1-\kappa}{\kappa-p_1} + 1 \right] \left[\tau_1 - \frac{1}{\kappa} \frac{d\kappa}{dA} \frac{p_1}{1-p_1} \right]$$

$$(2) \quad \delta_2 = \left[\frac{1-\kappa}{\kappa-p_1} + 1 \right] [\delta_1 + \Delta], \text{ where } \Delta(A) \text{ is a function on } [0, T_k] \text{ defined by}$$

$$(3) \quad \Delta(A) = \Delta_1(A) + \Delta_2(A) + \Delta_3(A), \text{ with}$$

$$\Delta_1 = \frac{1-\kappa}{\kappa-p_1} \tau_1^2, \quad \Delta_2 = -\frac{2\tau_1}{\kappa-p_1} \frac{d\kappa}{dA} + \frac{1}{\kappa^2} \frac{2\kappa-p_1}{\kappa-p_1} \frac{p_1}{1-p_1} \left[\frac{d\kappa}{dA} \right]^2, \quad \Delta_3 = -\frac{1}{\kappa} \frac{p_1}{1-p_1} \frac{d^2\kappa}{dA^2}.$$

Comments on lemma 4.5

- In the application of this theorem it is intended to substitute $\tau_2(t)$ by

¹⁾ time points A for which (1), (2) and (3) are not defined are excluded.

successively an unknown transmission function $\lambda(t)$, its spline approximation $\lambda_s(t)$ and the average trend function $\Lambda_s(t)$. As the spline approximations $\lambda_s(t)$ and $\Lambda_s(t)$ may become negative, $\tau_2(t)$ has been allowed to assume negative values.

- Without presupposing a past-specificity $\xi(A) = 1$, the function $\tau_1(t)$ will play the role of the antibody related transmission function $\lambda^a(t)$, its spline approximation $\lambda_s^a(t)$ and the average antibody related spline-trend $\Lambda_s^a(t)$.

Proof of lemma 4.5: From the definition of $p_1(A)$ and $p_2(A)$ it is easily derived that ($i=1,2$): $\tau_i(A) = \frac{d}{dA} [-e \log(1-p_i(A))]$. For a time point A with $p_2(A) \neq 0$ we have

$$p_1(A) = \kappa(A) p_2(A) \quad -\infty < \kappa(A) < \infty.$$

Theorem 4.2 and theorem 4.3 were based on the relations (4.16), (4.17) and (4.19). In the derivation of these theorema, however, the conditions that $p^a(A)$ and $p(A)$ are increasing and are strictly positive were not used. Furthermore it was not essential for these two theorema to hold that $n(A)$ is bounded. Hence we can formally substitute in formula (4.15), (4.22) and (4.23) the $\lambda(A)$, $\gamma(A)$, $\lambda^a(A)$, $\gamma^a(A)$ and $n(A)$ by $\tau_2(A)$, $\delta_2(A)$, $\tau_1(A)$, $\delta_1(A)$ and $\kappa(A)$ respectively. This completes the proof of lemma 4.5.

In the following only time points A are considered for which $0 < p_1(A) < 1$ and $0 < p_2(A) < 1$. Generalizations of lemma 4.1 and lemma 4.2 are then straight forward.

Lemma 4.6 let D be defined by $D = \{A | 0 < p_1(A) < 1 \text{ and } 0 < p_2(A) < 1\}$. Furthermore let for each $A \in D$, Δ_{\min} be defined by $\Delta_{\min} = \min \Delta(\kappa(A) \cdot \frac{d\kappa}{dA}, \frac{d^2\kappa}{dA^2})$. For $A \in D$ the following relations hold:

$$\text{sign } \delta_2 = \text{sign } (\delta_1 + \Delta), \text{ where } \Delta \geq \Delta_{\min} \text{ and} \quad (4.49)$$

$$\Delta_{\min} = - \frac{\kappa + \kappa p_1 - p_1}{2\kappa - p_1} \frac{\tau_1^2}{p_1} - \frac{1}{\kappa} \frac{d^2\kappa}{dA^2} \frac{p_1}{1-p_1}. \quad (4.50)$$

Proof From lemma 4.5 it follows that $\text{sign } \delta_2 = \text{sign} \left[\frac{1-\kappa}{\kappa-p_1} + 1 \right] \text{ sign } (\delta_1 + \Delta)$.

The term $\frac{1-\kappa}{\kappa-p_1} + 1$ is equal to $\frac{1-p_1}{\kappa-p_1}$. For $A \in D$ we have $0 < p_1 < 1$ and

$p_1 < \kappa < \infty$ and thus $\frac{1-\kappa}{\kappa-p_1} + 1$ is strictly positive. Hence it follows that

$\text{sign } \delta_2 = \text{sign } (\delta_1 + \Delta)$. As in theorem 4.3, the term Δ is called the *sign disturbance coefficient*.

To proof (4.50) an analogous way is followed as in lemma 4.1, section 4.3.2.

Let $r(A) = \frac{1}{\kappa(A)} \frac{d\kappa}{dA}$ with $p_1(A) < \kappa(A) < \infty$, then we have:

$$\Delta = ar^2 + br + c + d, \text{ where}$$

$$a = \frac{2\kappa - p_1}{\kappa - p_1} \frac{p_1}{1-p_1}, \quad b = - \frac{2\kappa}{\kappa - p_1} \tau_1, \quad c = \frac{1-\kappa}{\kappa - p_1} \tau_1^2, \quad d = - \frac{1}{\kappa} \frac{d^2\kappa}{dA^2} \frac{p_1}{1-p_1}.$$

As a function of r , Δ is a polynomial of degree 2. For a time point $A \in D$ the coefficient a is strictly positive. The sign disturbance coefficient assumes its minimum value if $r=r_{\min}$, with r_{\min} given by $r_{\min} = \frac{\kappa}{2\kappa - p_1} \frac{p_1}{1-p_1} \tau_1$. From the substitution of $r=r_{\min}$ in the formula of Δ it immediately follows that relation (4.50) holds.

Lemma 4.7 Let D be defined by $D = \{A \mid 0 < p_1(A) < 1 \text{ and } 0 < p_2(A) < 1\}$.

Furthermore let $A \in D$ for which $\kappa(A)$ is non-convex and $\delta_1(A) > 0$. Let the function $g(\kappa)$ be defined by (A fixed)

$$g(\kappa) = \delta_1 + \Delta_{\min} \quad (\delta_1 > 0; p_1 < \kappa < \infty) \quad (4.51)$$

$$\text{with } \Delta_{\min} = - \frac{\kappa + \kappa p_1 - p_1}{2\kappa - p_1} \frac{\tau_1^2}{p_1} - \frac{1}{\kappa} \frac{d^2\kappa}{dA^2} \frac{p_1}{1-p_1},$$

then we have for $A \in \{A | A \neq D \text{ and } \frac{d^2\kappa}{dA^2} \leq 0\}$:

(1) The function $g(\kappa)$ is strictly positive $\iff \delta_1 > \frac{1+p_1}{2p_1} \tau_1^2$

(2) let for δ_1 in interval $(\tau_1^2, \frac{1+p_1}{2p_1} \tau_1^2)$ the value κ_c be defined by

$$\kappa_c = \frac{\tau_1^2 - \delta_1 p_1}{(1+p_1)^{-1} \tau_1^2 - 2\delta_1}, \text{ then} \quad (4.52)$$

if $\kappa < \kappa_c$ then $g(\kappa) > 0$, and (4.53)

if $\kappa > \kappa_c$ then sign $g(\kappa)$ is uncertain.

Proof: The function $g(\kappa)$ is monotone decreasing for $p_1 < \kappa < \infty$. The values of $g(\kappa)$ in the "end points" of the interval are $g(p_1) = \delta_1 - \tau_1^2 - \frac{d^2\kappa}{dA^2} \frac{1}{1-p_1}$;
 $g(\infty) = \delta_1 - \frac{1+p_1}{2p_1} \tau_1^2$.

The function $g(\kappa)$ is strictly positive $\iff g(\infty) \geq 0 \iff \delta_1 \geq \frac{1+p_1}{2p_1} \tau_1^2$.

The function is strictly negative $\iff g(p_1) \leq 0 \iff \delta_1 \leq \tau_1^2 + \frac{d^2\kappa}{dA^2} \frac{1}{1-p_1}$.

The function $g(\kappa)$ takes on positive and negative values in the interval

$p_1 < \kappa < \infty \iff g(p_1) > 0 \text{ and } g(\infty) < 0 \iff \delta_1 > \tau_1^2 + \frac{d^2\kappa}{dA^2} \frac{1}{1-p_1}$ and
 $\delta_1 < \frac{1+p_1}{2p_1} \tau_1^2$.

Consider the function $g'(\kappa) = \delta_1 - \frac{\kappa + \kappa p_1 - p_1 \tau_1^2}{2\kappa - p_1} \frac{\tau_1^2}{p_1}$ ($p_1 < \kappa < \infty$).

The function $g'(\kappa)$ is monotone decreasing and assumes positive and negative values $\iff g'(p_1) > 0 \text{ and } g'(\infty) < 0 \iff \delta_1 > \tau_1^2$ and $\delta_1 < \frac{1+p_1}{2p_1} \tau_1^2$.

It is easily verified that $g'(\kappa_c) = 0$ with κ_c defined by (4.52).

As $g'(\kappa)$ is monotone decreasing it follows at once that

if $\kappa < \kappa^c$ then $g'(\kappa) > 0$
 if $\kappa > \kappa^c$ then $g'(\kappa) < 0$.

As $g(\kappa) \geq g'(\kappa)$ it is readily seen that (4.53) holds. This completes the proof of lemma 4.7.

Theorem 4.7 Let $\tau_1(t)$ and $\tau_2(t)$ be functions defined for $0 \leq t \leq T_k$, with continuous derivatives denoted by $\delta_1(t)$ and $\delta_2(t)$. T_k is an arbitrary positive value. Furthermore, let $p_1(A)$ and $p_2(A)$ be functions defined by

$$p_1(A) = \frac{1}{0} \exp \left(-\int_0^A \tau_1(t) dt \right) \text{ and } p_2(A) = \frac{1}{0} \exp \left(-\int_0^A \tau_2(t) dt \right), \text{ where}$$

$0 \leq A \leq T_k$ and k_1 and k_2 are constants, $0 < k_1 \leq 1$, $0 < k_2 \leq 1$.

Furthermore let $D = \{A \mid 0 < p_1(A) < 1 \text{ and } 0 < p_2(A) < 1\}$ and $\kappa(A) = \frac{p_1(A)}{p_2(A)}$ for $A \in D$.

Let $D_{\text{non-convex}} = \{A \mid A \in D, \frac{d^2 \kappa}{dA^2} \leq 0\}$. In order to interpret the course of $\tau_1(t)$ in a time point $A \in D_{\text{non-convex}}$ the following functions are used:

let $\ell(A)$, $u(A)$, $u^g(A)$ and $\kappa_c(A)$ be functions defined by¹⁾

$$\ell = \tau_1^2, \quad u = \frac{\tau_1^2}{p_1(2-p_1)}, \quad u^g = \frac{(1+p_1)\tau_1^2}{2p_1} \text{ and}$$

$$\kappa_c = \frac{\tau_1^2 - \delta_1 p_1}{(1+p_1^{-1}) \tau_1^2 - 2\delta_1}, \text{ where } \ell < \delta_1 < u^g.$$

For a time point $A \in D_{\text{non-convex}}$ with $\delta_1(A) > 0$ the following relations hold:

$$(1) \quad \text{If } \delta_1 \geq u^g \text{ then } \delta_2 > 0. \quad (4.54)$$

$$(2) \quad \text{If } \ell < \delta_1 < u^g \text{ then we have:} \quad (4.55)$$

if $\kappa < \kappa_c$ then $\delta_2 > 0$;

if $\kappa > \kappa_c$ then sign δ_2 is uncertain.

Furthermore for $A \in D_{\text{non-convex}}$ with $p_1(A) < \kappa(A) \leq 1$ the upperbound $u(A)$ instead of $u^g(A)$ can be used for determining the sign of δ_2 , i.e.:

¹⁾ note: if A is suppressed in $\ell(A)$ then the notation ℓ is used.

$$(3) \quad \text{If } \delta_1 > u \text{ then } \delta_2 > 0. \quad (4.56)$$

$$(4) \quad \text{If } \ell < \delta_1 \leq u \text{ then we have:} \quad (4.57)$$

if $\kappa < \kappa_c$ then $\delta_2 > 0$;

if $\kappa > \kappa_c$ then sign δ_2 is uncertain.

Proof: Using lemma 4.5, lemma 4.6 and lemma 4.7 we have:

$\text{sign } \delta_2 = \text{sign}(\delta_1 + \Delta)$, where $\Delta \geq \Delta_{\min}$ and the function $g(\kappa)$ is defined by

$g(\kappa) = \delta_1 + \Delta_{\min}$, where $\delta_1 > 0$ and $p_1 < \kappa < \infty$.

The relations (4.54) and (4.55) follow immediately from lemma 4.7. In order to prove (4.56) consider the critical value κ^c , this value κ^c has been defined for values δ_1 with $\ell < \delta_1 < u^g$. As a function of δ_1 , $\kappa^c(\delta_1)$ is monotone increasing. In the "end points" of the interval we have

$$\kappa_c(\ell) = p_1, \quad \kappa_c(u^g) = \infty.$$

Furthermore $\kappa_c(u) = 1$. Therefore for $A \in D_{\text{non-convex}}$ with $p_1(A) < \kappa(A) \leq 1$ it follows that: if $u < \delta_1 < u^g$ then $\kappa^c(\delta_1) > 1$ and thus $\kappa < \kappa^c$. Hence as a consequence of (4.55) it immediately follows that $\delta_2 > 0$ and thereby (4.56) holds. Relation (4.57) is a special case of (4.55). This completes the proof of theorem 4.7.

Remarks on the upperbound $u^g(A)$, $u(A)$

The (generalized) upperbound $u^g(A)$ is larger than the upperbound $u(A)$. The ratio $\frac{u^g(A)}{u(A)}$ assumes its maximum value for $p_1(A) = \frac{1}{2}$, furthermore it is readily verified that

$$u(A) < u^g(A) \leq \frac{9}{8} u(A), \quad (4.58)$$

hence the upperbound $u^g(A)$ is bounded by $u(A)$.

For $p_1(A)$ tends to 0 and $p_1(A)$ tends to 1, we have $\frac{u^g(A)}{u(A)} \rightarrow 1$. Furthermore for $p_1(A)$ near to 1, the upperbounds $u^g(A)$ and $u(A)$ approach the lowerbound $l(A)$,

i.e.: $u^g(A) \rightarrow 1(A)$ and $u(A) \rightarrow 1(A)$ for $p_1(A) \rightarrow 1$.

4.4.2.2 Relationship between $\lambda^a(t)$ and $\lambda(t)$

Theorem 4.7 is used for the interpretation of a decrease in $\lambda^a(t)$. A past-specificity $\xi(A) = 1$ nor a monotone increasing seropositivity rate $p^a(A)$ with age nor $\frac{d^2\eta}{dA^2} = 0$ is presupposed in the following. The ratio between the seropositivity rate $p^a(A)$ and the cumulative inoculation rate $p(A)$ is called the seropositivity-inoculation ratio (S-I ratio), this ratio is written as $\theta(A)$

$$\theta(A) = \frac{p^a(A)}{p(A)} \quad (p(A) \neq 0). \quad (4.59)$$

The S-I ratio is in a sense a generalization of the past-sensitivity $\eta(A)$; from (4.18) it follows that

$$\theta(A) = \eta(A) + [1 - \xi(A)] \frac{1 - p(A)}{p(A)}, \quad (4.60)$$

therefore in case the past-specificity $\xi(A) = 1$, we have

$$\theta(A) = \eta(A). \quad (4.61)$$

If $\xi(A) < 1$ and $p(A)$ is small then $\theta(A)$ may assume values > 1 .

From the definition of $\lambda(A)$ and $\lambda^a(A)$ it follows that (see (4.16) and (4.17))

$$p(A) = 1 - \exp \left[- \int_0^A \lambda(t) dt \right], \quad p^a(A) = 1 - (1 - p^a(0)) \exp \left[- \int_0^A \lambda^a(t) dt \right].$$

As $p(0) = 0$ it is easily seen from (4.18) that $p^a(0) = 1 - \xi(0)$. We are now in the position to apply theorem 4.7. The functions $\tau_1(t)$, $\tau_2(t)$, $p_1(A)$, $p_2(A)$, $\kappa(a)$ in this theorem are substituted by:

$$\tau_1(t) = \lambda^a(t), \quad p_1(A) = p^a(A), \quad \tau_2(t) = \lambda(t), \quad p_2(A) = p(A) \text{ and } \kappa(a) = \theta(A).$$

The constants k_1 and k_2 are chosen as $k_1 = 1 - p^a(0) = \xi(0)$ and $k_2 = 1$. Assuming the past-specificity $\xi(A) = 1$ or assuming $p^a(A) \leq p(A)$ it follows that

$$p^a(A) < \theta(A) \leq 1. \quad (4.62)$$

Let $l(A)$, $u(A)$, $u^g(A)$, $\theta_c(A)$ be functions defined by the right-hand side of the corresponding formulae in theorem 4.7 ($p_1(A)$, $\tau_1(A)$, $\delta(A)$ are substituted by respectively $p^a(A)$, $\gamma^a(A)$, $\gamma^a(A)$), then it is readily seen that the following theorem holds.

Theorem 4.8 If the S-I ratio $\theta(A)$ is non-convex in a time point A then:

(1) If $\gamma^a \geq u^g$ then $\gamma > 0$.

(2) If $l < \gamma^a < u^g$ then:

if $\theta < \theta_c$ then $\gamma > 0$;

if $\theta > \theta_c$ then sign γ is uncertain.

Furthermore if the past-specificity $\xi(A) = 1$ or if $p^a(A) \leq p(A)$ the upperbound $u(A)$ can be used instead of $u^g(A)$ for the determination of sign γ , i.e.

(3) If $\gamma^a > u$ then $\gamma > 0$.

(4) If $l < \gamma^a \leq u$ then:

if $\theta < \theta_c$ then $\gamma > 0$;

if $\theta > \theta_c$ then sign γ is uncertain.

Remarks regarding the function $\theta_c(A)$

The function $\theta_c(A)$ is called the critical S-I ratio. The critical S-I ratio is an extension of the critical past-sensitivity $\eta_c(A)$. The function $\theta_c(A)$ is defined for $\gamma^a(A)$ in the interval $l(A) < \gamma^a(A) < u^g(A)$, whereas $\eta_c(A)$ has been defined for $l(A) < \gamma^a(A) \leq u(A)$. However, in the interval $\{A | l(A) < \gamma^a(A) \leq u(A)\}$ $\theta_c(A)$ and $\eta_c(A)$ assume the same values

$$\theta_c(A) = \eta_c(A). \quad (4.63)$$

Hence theorem 4.8 can be considered a generalization of theorem 4.5, the function $u^g(A)$ is called the generalized upperbound.

4.4.2.3 Relationship between $\lambda_s^a(t)$ and $\lambda_s(t)$

The spline approximations $\lambda_s(t)$ and $\lambda_s^a(t)$ are defined by (see (2.4))

$$\lambda_s(t) = \frac{d}{dt} v_s(t) \quad \lambda_s^a(t) = \frac{d}{dt} v_s^a(t).$$

The functions $v_s(t)$ and $v_s^a(t)$ are the natural cubic spline approximations of $v(t)$ respectively $v^a(t)$. The functions $v(t)$ and $v^a(t)$ are given by ¹⁾

$$v(A) = -e \log(1-p(A)) \quad v^a(A) = -e \log(1-p^a(A)). \quad (4.64)$$

The junction points of the natural cubic spline approximations are at time points $T_j = A_j$ ($j=0, 1, \dots, k$). Hence $v_s(t)$ and $v_s^a(t)$ passes through the plane points $(A_j, v(A_j))$ ($j=0, 1, \dots, k$) and $(A_j, v^a(A_j))$ ($j=0, 1, \dots, k$) respectively. The spline approximation $p_s^a(A)$ of the seropositivity rate $p^a(A)$ is defined in a similar way as the spline approximation $p_s(A)$ of the cumulative inoculation rate $p(A)$ (see Appendix 2, (2.8))

$$p_s(A) = \frac{-v_s(A)}{1-e} \quad p_s^a(A) = \frac{-v_s^a(A)}{1-e}. \quad (4.65)$$

In the knots A_j ($j=0, 1, \dots, k$) we have (4.66)

$$\begin{aligned} p_s(0) &= p(0) = 0 & p_s^a(0) &= p^a(0) = 1-\xi(0) \\ p_s(A_j) &= p(A_j) & p_s^a(A_j) &= p^a(A_j). \end{aligned}$$

The ratio between $p_s^a(A)$ and $p_s(A)$ is called spline approximation of the seropositivity-inoculation ratio and this ratio is written as $\theta_s(A)$

$$\theta_s(A) = \frac{p_s^a(A)}{p_s(A)} \quad (p_s(A) \neq 0). \quad (4.67)$$

In the knots A_j ($j=1, \dots, k$) $\theta_s(A)$ assumes the same values as $\theta(A)$

$$\theta_s(A_j) = \theta(A_j) \quad (j=1, \dots, k). \quad (4.68)$$

¹⁾ Note that: $v(A_0) = v(0) = 0$ and $v^a(A_0) = v^a(0) = -e \log(1-p^a(0)) = -e \log \xi(0)$.

If the past-specificity $\xi(A)$ of the serological test is 1 then it follows from (4.61) and (4.68) that

$$\theta_s(A_j) = \eta(A_j) \quad (j=1, \dots, k). \quad (4.69)$$

In this situation $\theta_s(A)$ is called the spline approximation of the past-sensitivity $\eta(A)$ and the notation $\eta_s(A)$ is used. From the definition of $p_s(A)$ and $p_s^a(A)$ it follows that

$$p_s(A) = 1 - \exp \left[- \int_0^A \lambda_s(t) dt \right] \quad p_s^a(A) = 1 - \xi(0) \exp \left[- \int_0^A \lambda_s^a(t) dt \right]. \quad (4.70)$$

We are now in the position to apply theorem 4.7. Let $l_s(A)$, $u_s^g(A)$, $u_s^g(A)$ and $\theta_s^c(A)$ be functions defined by

$$l_s = [\lambda_s^a]^2, \quad u_s = \frac{[\lambda_s^a]^2}{p_s^a(2-p_s^a)}, \quad u_s^g = \frac{1+p_s^a}{2p_s^a} [\lambda_s^a]^2 \text{ and} \quad (4.71)$$

$$\theta_s^c = \frac{[\lambda_s^a]^2 - \gamma_s^a p_s^a}{(1+(p_s^a)^{-1}) [\lambda_s^a]^2 - 2\gamma_s^a}, \text{ where } l_s < \gamma_s^a < u_s^g. \quad (4.72)$$

The function $l_s(A)$, $u_s(A)$, $u_s^g(A)$ and $\theta_s^c(A)$ are called spline approximations of the lowerbound $l(A)$, the upperbound $u(A)$, the generalized upperbound $u^g(A)$ and the critical seropositive-inoculation ratio $\theta_c(A)$ respectively¹⁾.

The analogon of theorem 4.8 in terms of spline approximation is presented in theorem 4.9.

Theorem 4.9 If the spline approximation $\theta_s(A)$ of the S-I ratio $\theta(A)$ is non-convex in a time point A and the spline approximations $p_s^a(A)$ and $p_s(A)$ are positive then:

(1) If $\gamma_s^a > u_s^g$ then $\gamma_s > 0$.

(2) If $l_s \leq \gamma_s^a < u_s^g$ then:

¹⁾ note that the spline approximation of $\theta_c(A)$ is written as $\theta_s^c(A)$.

if $\theta_s < \theta_s^c$ then $\gamma_s > 0$;
 if $\theta_s > \theta_s^c$ then sign γ_s is uncertain.

Furthermore if the past-specificity $\xi(A) = 1$ or $p^a(A) \leq p(A)$ we can use in the knots A_j ($j=1, \dots, k$) the upperbound $u_s(A)$ for the determination of sign $\gamma_s(A)$.

Comment on the spline approximation of the critical S-I ratio

If the past-specificity $\xi(A)$ of the serological test is 1 then $\theta_s(A) = n_s(A)$ and accordingly $\theta_s^c(A)$ is called the spline approximation of the critical past-sensitivity and the notation $n_s^c(A)$ is used.

4.4.2.4 Relationship between $\Lambda_s^a(t)$ and $\Lambda_s(t)$

In a non-homogeneous population which consists of m homogeneous sub-populations, the average seropositivity rate $P^a(A)$ and the average antibody related transmission function $\Lambda^a(t)$ are defined in a similar way as $P(A)$ and $\Lambda(t)$ (see Appendix 3, (3.1) and (3.4)), thus

$$P^a(A) = 1 - \prod_{i=1}^m (1-p_i^a(A))^{w_i} \quad \Lambda^a(t) = \sum_{i=1}^m w_i \lambda_i^a(t). \quad (4.73)$$

Analogously to the definition of $P_s(A)$ and $\Lambda_s(t)$ the spline approximations of these functions are defined by

$$P_s^a(A) = 1 - \prod_{i=1}^m (1-p_{i,s}^a(A))^{w_i} \quad \Lambda_s^a(A) = \sum_{i=1}^m w_i \lambda_{i,s}^a(t). \quad (4.74)$$

The rate of change in $\Lambda^a(t)$ and $\Lambda_s^a(t)$ is denoted by $\Gamma^a(t)$ and $\Gamma_s^a(t)$ respectively. The ratio between the average seropositivity rate $P^a(A)$ and the average cumulative inoculation rate $P(A)$ is called average S-I ratio and this ratio is written as $\Theta(A)$

$$\Omega(A) = \frac{P^A(A)}{P(A)} \quad (P(A) \neq 0). \quad (4.75)$$

If the past-specificity $\xi(A)$ of the serological test is 1 or $p_i^A(A) \leq p_i(A)$ ($i=1, \dots, m$) then we have $\Omega(A) \leq 1$. In case $\xi(A) = 1$ we call $\Omega(A)$ the *average past-sensitivity of the serological test* and use the notation $H(A)$. The ratio between $P_s^A(A)$ and $P_s(A)$ is called spline approximation of the average S-I ratio and is notated as $\Theta_s(A)$

$$\Theta_s(A) = \frac{P_s^A(A)}{P_s(A)} \quad (P_s(A) \neq 0). \quad (4.76)$$

In the knots A_j ($j=1, \dots, k$) the function Θ_s assumes the same values as the function Ω

$$\Theta_s(A_j) = \Omega(A_j) \quad (j=1, \dots, k). \quad (4.77)$$

If the past-sensitivity $\xi(A)$ of the serological test is 1 then $\Theta_s(A)$ is called the spline approximation of the average past-sensitivity $H(A)$ and the notation $H_s(A)$ is used. In the knots we then have

$$H_s(A_j) \leq 1 \quad (j=1, \dots, k). \quad (4.78)$$

From the definition of $P_s(A)$ and $P_s^A(A)$ it follows that

$$P_s(A) = 1 - \exp \left[\int_0^A \lambda_s^A(t) dt \right] \text{ and } P_s^A(A) = 1 - \xi(0) \exp \left[- \int_0^A \lambda_s^A(t) dt \right]. \quad (4.79)$$

Now we are again in the position to apply theorem 4.7. Let $L_s(A)$, $U_s(A)$, $U_s^g(A)$ and $\Theta_s^c(A)$ be functions defined by the right-hand sides of the corresponding formulae in (4.71) and (4.72) in which p_s^A , λ_s^A , γ_s^A are substituted by respectively P_s^A , λ_s^A , γ_s^A . The function $\Theta_s^c(A)$ is called spline approximation of the average critical S-I ratio. From theorem 4.7 immediately follows the analogon of theorem 4.9 for a non-homogeneous population.

Theorem 4.1.6 If the spline approximation $\Omega_s^c(A)$ of the average S-I ratio

$\Omega(A)$ is non-convex in a time point A and the spline approximations $P_s^a(A)$ and $P_s^c(A)$ are positive then:

(1) If $P_s^a > U_s^g$ then $\Omega_s^c > 0$.

(2) If $L_s < P_s^a < U_s^g$ then:

if $\Omega_s < \Omega_s^c$ then $\Omega_s^c > 0$;

if $\Omega_s > \Omega_s^c$ then sign Ω_s^c is uncertain.

Furthermore if the past-specificity $\xi(A) = 1$ or $p_i^a(A) \leq p(A)$ ($i=1, \dots, m$) we can use in the knot, A_j ($j=1, \dots, k$) the upperbound $U_s(A)$ instead of $U_s^g(A)$ for the determination of the sign of $\Omega_s^c(A)$.

Comment on the spline approximation of the average critical S-I ratio

If the past-specificity $\xi(A)$ of the serological test is 1 then $\Omega_s^c(A)$ is called the spline approximation of the average critical past-sensitivity and the notation $H_s^c(A)$ is used.

REFERENCES

- Hilberg, J.H., Nilson, E.N. and Walsh, J.L. (1967). *The theory of splines and their applications*. New York, Academic Press.
- Anscombe, F.J. (1956). On estimating binomial response relations. *Biometrika*, 43: 461-464.
- Bailey, N.T.J. (1975). *The mathematical theory of infectious diseases*. London, Charles Griffin.
- Bartlett, M.S. (1956). Deterministic and stochastic models for recurrent epidemics. *Proc. 3rd Berkeley Symp. Math. Statist. Prob.*, 4: 81-109.
- Bartlett, M.S. (1960). *Stochastic population models in ecology and epidemiology*. London, Methuen.
- Brögger, R.C., Mathews, H.M., Storey, J., Ashkar, T.S., Brögger, S. and Molineaux, L. (1978). Changing patterns in the humoral immune response to malaria before, during, and after the application of control measures: a longitudinal study in the West African Savanna. *Bull. Wld Hlth Org.*, 56: 579-600.
- Bruce-Chwatt, L.J., Draper, C.C., Konfortion, P. (1973). Sero-epidemiological evidence of eradication of malaria from Mauritius. *The Lancet*, 8: 547-551.
- Bruce-Chwatt, L.J. (1976). Mathematical models in the epidemiology and control of malaria. *Trop. geogr. Med.*, 28: 1-8.
- Bruce-Chwatt, L.J. (1980). *Essential malariology*. London, Heinemann.
- Cohen, J.E. (1973). Heterologous immunity in human malaria. *The quarterly review of biology*, 48: 467-489.
- Cook, G.W., Kerridge, D.F. and Tryce, J.D. (1974). Estimations of functions of a binomial parameter. *Sankhyā*, 36(A): 443-448.
- Cox, D.R. and Lewis, P.A.W. (1966). *The statistical analysis of series of events*. London, Methuen.
- Cox, D.R. (1970). *The analysis of binary data*. London, Methuen.
- Dietz, K., Molineaux, L. and Thomas, A. (1974). A malaria model tested in the African Savannah. *Bull. Wld Hlth Org.*, 50: 347-357.
- Dietz, K. (1976). in *Mathematical models in medicine*. (ed. Levin, S.A.). New York, Springer.
- Dietz, K., Molineaux, L. and Thomas, A. (1978). Further epidemiological evaluation of a malaria model. *Bull. Wld Hlth Org.*, 56: 565-571.
- Dowling, M.A.C., Rickman, L.R., Shute, J.T., Menzies, N.G.F. (1966). A field method of blood concentration for the improved diagnosis of scanty parasitaemias in malaria practice. WHO/MAL 66.535.

- Brayner, C.C., Miller, A. and Carpenter, P.G. (1979). The epidemiologic interpretation of serologic data in malaria. Am. J. Trop. Med. and Hyg., 21: 696-703.
- Feveritt, B.J. (1977). The analysis of contingency tables. New York-London, John Wiley.
- Fine, P.L.C. (1975). Superinfection - a problem in formulating a problem (an historical critique of Macdonald's theory). Tropical diseases bulletin, 72: 475-488.
- Fisher, R.A. (1947). Statistical methods for research workers. Edinburgh, Oliver and Boyd.
- Garrett-Jones, C. (1961). The human blood index of malaria vectors in relation to epidemiological assessment. Bull. Wld Hlth Org., 30: 241-261.
- Giglioli, G. (1951). Eradication of Anopheles darlingi from the inhabited areas of British Guyana by DDT residual spraying. J. Nat. Mal. Soc., 10: 142-.
- Hald, A. (1957). Statistical theory with engineering applications. New York-London, John Wiley.
- Haldane, J.B.S. (1908). The estimation and significance of the logarithm of a ratio of frequencies. Annals of Human Genetics, 20: 309-311.
- Haldane, J.B.S. (1955). Almost unbiased estimates of functions of frequencies. Sankhyā, 17: 201-208.
- Kaay, R.J. van der (1970). Malaria in Surinam, a sero-epidemiological study. Meppel, The Netherlands, Krips Repro B.V. (thesis).
- Khinchin, A.I. (1958). On Poisson sequences of chance events. Theory of probability and its applications (English translation of the Soviet Journal), 1: 291-297.
- Lafaye, A. (1970). Un modèle exponentiel simple, la fonction catalytique en épidémiométrie tuberculinique. Bull. Organ. Mond. Santé, 54: 633-643.
- Lane, J. (1945). Anophelines of the Neotropical region. In: Mariology, Vol. I, chapter 17. Ed. Boyd, M.F. Philadelphia, W.B. Saunders Comp.
- Tee, E.T. (1980). Statistical methods for survival data analysis. Belmont, California, Lifetime learning publications.
- Lobel, R.J., Nájera, F.J., N'ien, Wm., Muñoz, P. and Mathews, H.M. (1976). Seroepidemiologic investigations of malaria in Guyana. J. Trop. Med. Hyg., 79: 275-284.
- Macdonald, G. (1950). The analysis of infection rates in diseases in which superinfection occurs. Tropical diseases bulletin, 47: 907-915.
- Macdonald, G. (1957). The epidemiology and control of malaria. London, Oxford Univ. Press.
- May, P.M. and Andreon, H.M. (1973). Population biology of infectious diseases: part II. Nature, 280: 455-461.
- Meuwissen, J.H.P.H. (1974). The indirect haemagglutination test for malaria and its application to epidemiological surveillance. Bull. Wld Hlth Org., 50: 277-286.
- Molineaux, L., Storey, J., Cohen, J. and Thomas, A. (1977). Longitudinal study of *P.falciparum*, *P.malariae* and *P.ovale*, in the West African Savanna, in the absence of control measures: II. Relationships between the species of plasmodium, in particular *P.falciparum* and *P.malariae*. Unpublished document MPD-012/78.17.
- Molineaux, L. and Gramiccia, G. (1980). The Garki project. Research on the epidemiology and control of malaria in the Sudan Savanna of West Africa. Geneva, World Health Organization.
- Mood, A.M., Graybill, F.A., Boes, D.C. (1974). Introduction to the theory of statistics. London, McGraw-Hill.

- Murch, H. (1963). Catalytic models in epidemiology. Cambridge, Massachusetts, Harvard Univ. Press.
- Tansey, G. (1930, 1963 'nd edition). A textbook of malaria eradication. London, Oxford Univ. Press.
- Parzen, I. (1970). Stochastic Processes. San Francisco, Holden-Day.
- Peston, S. (1967). Quantitative research in human biology and medicine. Bristol.
- Radcliffe, J. (1974). The periodicity of endemic malaria. *J. Appl. Prob.*, 11: 562-567.
- Ross, K. (1911). The prevention of malaria. London, Murray.
- Flinck, C.H. L. and van Eeden, C. (1951). Statistiek voor medici. Leiden, The Netherlands, Stafleu.
- Schwarz, O., Dietz, K. and Fröner, G. (1973). Antibody against hepatitis A in seven european countries. II Statistical analysis of cross-sectional surveys. *Am. J. Epidemiol.*, 110: 70-76.
- Snijders, D.J. (1975). Random point processes. New York, John Wiley.
- Swinton, J. (1966). Statistical methods in malaria eradication. Geneva, Wld Hlth Org.
- Obre, J.E., Coatney, G.R. (1961). Fluorescent antibody staining of human malaria parasites. *Exp. Parasitology*, 11: 128-132.
- Vollen, A., C'Neil, P. (1971). Immunofluorescence method suitable for large-scale application to malaria. *Bull. WHO*, 45: 524-529.
- World Health Organization (1968). Immunology of malaria. Techn. Rep. Series no. 346, Geneva.
- World Health Organization (1975). Developments of malaria immunology. Techn. Rep. Series no. 579, Geneva.
- Yorke, J.A. and London, W.P. (1975). Recurrent outbreaks of measles, chickenpox and mumps. II: Systematic differences in contact rates and stochastic effects. *Amer. J. Epidemiol.*, 98: 469-482.

Some additional mathematical references not mentioned in the text

- Boor, C. de (1978). A practical guide to splines. New York, Springer-Verlag.
- Buse, A. and Lim, T. (1977). Cubic splines as a special case of restricted least squares. *JASA*, 72: 64-68.
- Clevenson, M.L. (1977). Bayes linear estimators of the intensity function of the nonstationary Poisson process. *JASA*, 72: 112-120.
- Ertel, J.E. and Fowlkes, E.B. (1976). Some algorithms for linear spline and piecewise multiple linear regression. *JASA*, 71: 640-648.
- Greville, T.N.E. (1969). Theory and applications of spline functions. New York, Academic Press.
- Lewis, P.A.W. (1979). Recent results in the statistical analysis of univariate point processes. In *Stochastic Point Processes*, Ed. P.A.W. Lewis, 1-54. New York, John Wiley.
- Poirier, D.J. (1973). Piecewise regression using cubic splines. *JASA*, 68: 515-524.
- Reinsch, C.H. (1967). Smoothing by spline functions. *Numerische Mathematik*, 10: 177-183.
- Schoenberg, I.J. (1969). Approximations with special emphasis on spline functions. New York, Academic Press.
- Wold, S.W. (1974). Spline functions in data analysis. *Technometrics*, 16: 1-11.
- Wright, I.W., Wegman, E.J. (1980). Isotonic, convex and related splines. *The Annals of Statistics*, 8: 1023-1035.

AUTHOR INDEX

- Ahlberg, J.H., 255, 257, 260, 262
Anderson, R.M., 224
Anscombe, F.J., 269
Ashkar, T.S., 198, 203, 205
Bailey, N.T.J., 6, 223
Bartlett, M.S., 223
Boes, D.C., 266
Brögger, R.C., 198, 203, 205
Brögger, S., 198, 203, 205
Bruce-Chwatt, L.J., 6, 19, 40-42,
230, 231
Carpenter, R.G., 6, 19, 109, 161,
166, 168, 226, 233
Ch'en, Wan I., 160-168, 226
Coatney, G.R., 1
Cohen, J.E., 224, 225
Cook, G.W., 269, 270
Cox, D.R., 249, 267
Dietz, K., 2, 30, 224, 230¹⁾, 232
Dowling, M.A.C., 168
Draper, C.C., 6, 19, 109, 161, 166,
168, 226, 233
Eeden, C., 94
Everitt, B.S., 47
Fine, P.E.M., 232
Fisher, R.A., 66
Frösner, G.G., 30
Garrett-Jones, C., 5, 231
Giglioli, G., 165
Gramiccia, G., 224, 225, 232
Graybill, F.A., 266
Hald, A., 67
Haldane, J.B.S., 269
Kaay, H.J. van der, 168-184, 226
Kerridge, D.F., 269, 270
Khinchin, A.I., 251
Lafaye, A., 30
Lane, J., 225
Lee, E.T., 236
Lewis, P.A.W., 249
Lobel, H.O., 160-168, 226
London, W.P., 224
Macdonald, G., 5, 31, 35, 221, 222,
226, 231, 232
Mathews, H.M., 160-168, 198, 203,
205, 226
May, R.M., 224
Menzies, N.G.F., 168
Meuwissen, J.H.E.Th., 235
Molineaux, L., 6, 1971), 198, 203,
205, 224, 225, 227¹⁾, 232
Mood, A.M., 266
Muensch, H., 2, 3, 6, 30, 33, 233
Munroe, P., 160-168, 226
Najéra, A.J., 160-168, 205¹⁾, 206¹⁾,
226, 227¹⁾
Nilson, E.N., 255, 257, 260, 262
O'Neill, P., 77
Pampana, E., 222, 225, 230
Parzen, E., 249, 251
Peller, S., 230
Pryce, J.D., 260, 270
Ponnudurai, T., 75¹⁾
Radcliffe, J., 223
Rajakulendran, S., 75¹⁾
Rickman, L.R., 168
Ross, R., 5
Rümke, Chr.L., 94

- Schenzle, D., 30
Shute, J.T., 168
Snijder, D.L., 249, 251
Storey, J., 198, 203, 205,
 224, 225
Subramaniam, K., 75¹⁾
Swaroop, S., 32, 230
- Thomas, A., 6, 224, 225, 232
Tobie, J.E., 1
Voller, A., 6, 19, 77, 109, 161, 166,
 168, 226, 233
Walsh, J.L., 255, 257, 260, 262
World Health Organization, 1
Yorke, J.A., 224

¹⁾ indicated page does not refer to a reference

CURRICULUM VITAE

De auteur van dit proefschrift werd geboren op 28 maart 1944 te Arnhem. In 1960 behaalde hij het diploma MULO A en B. Na in 1962 het eindexamen HBS-B te hebben behaald aan de Lorentz-Scholengemeenschap in zijn geboorteplaats, begon hij de studie wiskunde aan de Katholieke Universiteit Nijmegen. In januari 1966 werd het kandidaatsexamen afgelegd met de bijvakken natuurkunde en sterrenkunde. Na twee student-assistentschappen te hebben vervuld -algebra en waarschijnlijkheidsrekening - werd op 5 juni 1969 het doctoraal examen wiskunde afgelegd met als hoofdvak analyse en bijvak waarschijnlijkheidsrekening en statistiek.

In het cursus jaar 1969-1970 was hij als leraar wiskunde werkzaam aan de Lorentz-Scholengemeenschap in Arnhem. Vanaf augustus 1970 is hij in dienst van de Katholieke Universiteit Nijmegen bij de Mathematisch Statistische Adviesafdeling (MSA), vanaf 1979 is hij als wetenschappelijk hoofdambtenaar aan deze afdeling verbonden. In de jaren 1973 en 1976-1980 doceerde hij bovendien lineaire modellen in het kader van de opleiding "statisticus VVS" (Vereniging Voor Statistiek).

In 1976 werd hem een plaats toegekend in de Universitaire Onderzoekspool; de doelstelling van het onderzoek was de ontwikkeling van een mathematisch-statistisch model voor de retrospectieve analyse van malaria transmissie op grond van transversaal serologisch bevolkingsonderzoek. De resultaten van die studie zijn neergelegd in het onderhavige proefschrift.

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STELLINGEN

I

Informatie omtrent de transmissie intensiteit in een homogene populatie ligt besloten in een aan de leeftijd gerelateerde parameter voor de frequentie van inoculaties. Dit impliceert dat het epidemiologisch verloop van malaria transmissie in principe kan worden gereconstrueerd op grond van transversaal bevolkingsonderzoek.

II

All exact science is dominated by the idea of approximation.

Bertrand Russell

III

Spline approximaties van de transmissie intensiteit, zoals gedefinieerd in deze studie, bezitten de eigenschappen "behoud van oppervlakte en maximale gladheid"; iedere spline approximatie beschrijft bepaalde aspecten van de onbekende transmissie intensiteit. Deze eigenschappen zijn uitermate gunstig voor de bestudering van alle type veranderingen in het verloop daarvan.

IV

Polynomials are wonderful even after they are cut into pieces.

I.J. Schoenberg

Voor de bestudering van het epidemiologisch verloop van de transmissie intensiteit in een niet homogene populatie kan deze populatie worden beschouwd als homogeen, als gebruik wordt gemaakt van een gewogen meetkundig gemiddelde van aan de leeftijd gerelateerde parameters voor de frequentie van inoculaties in homogene deel-populaties.

VI

In het algemeen is de "past-sensitivity" van een serologische test als functie van de leeftijd onbekend en bovendien verstrengeld met de transmissie intensiteit; als de onbekende past-sensitivity echter lineair wordt benaderd is het in principe mogelijk een afname in het verloop van malaria transmissie te detecteren op grond van een transversaal serologisch profiel van de bevolking.

VII

De in deze studie ontwikkelde methoden zijn in principe toepasbaar bij onderzoek naar het verloop van: besmettingskansen bij infectie ziekten, de "hazard rate" in overlevingsduur studies en de intensiteit van stochastische punt processen.

VIII

Toegepaste statistiek is gebaseerd op twee pijlers: model keuze en statistische analyse binnen het gekozen model. In het algemeen wordt te weinig aandacht besteed aan de kwantificering van foutenbronnen die betrekking hebben op de model keuze. De betrouwbaarheid van statistische uitspraken in praktische toepassingen wordt derhalve vaak overschat.

Bij een ontbreken van fundamenteel inzicht in de aard van een te bestuderen verschijnsel wordt vaak de aanduiding toevalsverschijnsel gehanteerd. De kans dat een toevalsverschijnsel wezenlijk op toeval berust is derhalve niet te schatten.

X

Laat hij die zoekt, niet ophouden te zoeken totdat hij vindt, en wanneer hij vindt zal hij in de war gebracht worden, en wanneer hij in de war is geweest, zal hij zich verwonderen - - -.

Uit het Evangelie volgens Thomas.

XI

De hoogste menselijke bestaansvorm lijkt als voorwaarde te hebben dat het "ik" zichzelf verliest, niet uit zwakte, maar bewust loslatend en leeg wordend.

Han Fortmann,
Oosterse Renaissance,
Ambo (1970), 59-60.

XII

De groei van de goeroe-bewegingen in het Westen weerspiegelt de toegenomen behoefte aan meditatie en mystiek. Het is te betreuren dat de kerken de positieve elementen in deze ontwikkeling te weinig onderkennen.

XIII

Grote aantallen respondenten, grote aantallen variabelen en uitgebreide statistische analyses met behulp van de computer beïnvloeden de betekenis van "significant $p \leq 0.05$ " in negatieve zin. Het is derhalve niet uitgesloten dat de betrouwbaarheid van "significant $p \leq 0.05$ " een dalende functie is van de tijd; dit impliceert dat recente medisch-statistische literatuur mogelijk onbetrouwbaarder is dan vergelijkbare literatuur van enkele tientallen jaren geleden.

XIV

De betrouwbaarheid van een proefschrift wordt verhoogd door de onbetrouwbare onderdelen ervan sterk te benadrukken.

J.A.M. van Druten,

Nijmegen, 1 october 1981.

