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Prediction of the flight of *Hyalesthes obsoletus*, vector of stolbur phytoplasma, using temperature sums

M. Maixner and M. Langer
BBA, Institute for Plant Protection in Viticulture, D-54470 Bernkastel-Kues, Germany - M.Maixner@BBA.de

Abstract: A method to predict the flight of the planthopper *Hyalesthes obsoletus*, the vector of stolbur-phytoplasma that causes Bois noir disease in grapevine, is presented. The parameters required for the calculation of temperature sums were determined by multiple calculations of temperature sums till the observed dates of the start of flight activity of adult planthoppers in different years and the identification of combinations of starting dates and temperature thresholds for day-degree accumulation that led to a minimum relative standard error. The method can be currently used for populations of *H. obsoletus* that live on bindweed (*Convolvulus arvensis*), only. It needs to be adjusted for populations on nettle (*Urtica dioica*) that show a different flight activity. The procedure described here could be useful for other insects, too, where relevant data of the life cycle cannot be determined in laboratory rearing.

Key words: *Hyalesthes obsoletus*, Bois noir, temperature sums, prediction of flight.

Introduction

The planthopper *Hyalesthes obsoletus* Signoret is the vector of stolbur-phytoplasma that, beside other diseases, causes Bois noir (BN) disease in grapevine (Maixner, 1994; Sforza et al., 1998; Alma et al., 2005). This xerothermic species reaches the northern border of its range in Germany where it depends on areas characterized by a favorable microclimate such as vineyards on the slopes of major rivers such as Rhine and Mosel. BN is of particular significance at such locations.

H. obsoletus is a univoltine and polyphagous species which feeds on a variety of herbaceous plants (Langer et al., 2003). Eggs are deposited on some of those plants where all larval instars feed on the roots. Third instar larvae move downward in the soil and hibernate in a depth where they are likely to be protected from frost. In spring they ascend to just below the soil surface where they remain until moulding of fifth instar nymphs to adults. Only these leave the soil and feed on shoots and leaves. Although grapevine is considered to be an accidental host only (Alma et al., 1987) it is occasionally approached by the adult plant-hoppers and can be inoculated with the phytoplasma by erroneous feeding. Temperature has an obvious effect on the development of *H. obsoletus* since the time of the emergence of adult vectors varies within approximately four weeks depending on weather conditions. The flight lasts for roughly seven weeks after the first occurrence of adults.

The BN phytoplasma depends on a cyclic change between plant and insect hosts since it is not transmitted vertically either in the insect or in the plant hosts. Different natural epidemiological cycles that include diverse host plant species exist in the field and both the vector populations and the strains of the pathogen appear to be adapted to the particular host plant species (Langer & Maixner, 2004). *Convolvulus arvensis* (bindweed) and *Urtica dioica* (nettle) are the most important host species for both, the vector and the phytoplasma. They are of variable significance in different geographic regions. While *C. arvensis* is the predominant host in Germany (Langer & Maixner, 2004) nettle is preferred in Italy for example (Alma et al., 1987).

Knowledge of the start, duration and intensity of the flight activity of *H. obsoletus* is useful where: (1) the occurrence of this species in a particular area needs to be confirmed; (2) sufficient numbers of vectors need to be sampled in order to determine the infestation of field populations as a measure of infection pressure of BN phytoplasma on grapevine; (3) growers need information at what time weed control should be omitted in order to prevent an intensified movement of vectors from weeds to grapevine. Therefore, a method was developed that allows determining the beginning of the flight activity of *H. obsoletus* using temperature data. We were obliged to use field data since breeding of the soil inhabiting and univoltine *H. obsoletus* in the laboratory is not efficient.

Methods

The parameters used for the calculation of temperature sums are listed in table 1. Yellow sticky traps were exposed in vineyards and adjacent fallow areas at Bernkastel-Kues, Mosel, in the years 1993 to 1996 and 1999 to 2004 from the mid of May before the usual flight activity until no vectors were caught anymore. The traps were removed weekly and checked for adult *H. obsoletus*. From 1999 to 2004 additional daily sweep net surveys were carried out from end of May until the first adult planthoppers were caught.

The daily average of the air temperature (AT) and the daily minimum of the soil surface temperature (ST) were calculated from the data recorded by a nearby weather station in a distance of approximately 700 m from the experimental plot on the opposite side of the Mosel River.

To find out the most suitable combination of starting date and threshold temperature, the temperature sums were calculated for all possible combinations of the starting date that varied from January 1 to April 30 and of the threshold temperature that was increased from 0 °C to 10 °C in steps of 0.1 °C. The total degrees above 0 °C were added to the sums for such days where the threshold temperature was reached or exceeded. The day-degree (dd) sums of AT and ST were accumulated separately for each year until the date of observed start of flight, and the mean, variance, standard deviation (SD) and relative standard error (RSE=standard

Table 1. Parameters used for the calculation of temperature sums. The optimal parameter sets were selected from a total of 48 000 combinations (2 measured temperatures x 2 observed starting dates of flight x 100 threshold temperatures x 120 starting dates of temperature accumulation).

Parameter	No of variations	Variation
Measured temperatures	2	Daily average of air temperature (AT) Daily minimum of soil surface temperature (ST)
Observed start of flight	2	First day and forth day of the one week trap exposure period
Threshold temperature	100	0 °C to 10 °C; dT=0.1 °C
Starting date of temperature accumulation	120	January 1 to April 30; dt=1 day

error/mean*100) of the temperature sums calculated from the different years where estimated. Certain combinations of starting date and threshold temperature were identified where RSE reached a minimum. In addition, best fitting temperature sums were also calculated for 'convenient starting dates' such as beginning or mid of a month. Day-degree sums till flight were determined in this way with the first (begin) and the forth day (middle), respectively, of the one week sticky trap exposure period considered to be the observed start of flight. The observed flight data were then compared to the dates predicted by the temperature sums that were calculated with the different sets of parameters and the average divergence between observed and predicted dates was determined.

In order to get information about the seasonal movement of *H. obsoletus* in the soil, larvae were dug monthly during the winter of 2003/2004 on the roots of *Ranunculus sp.* in a vineyard. This host plant was chosen because high population densities of the planthopper are often found where it is a predominant weed, although it is no host of the BN phytoplasma. We have no evidence that flight on *Ranunculus* differs from that on *Convolvulus*. Sampling at each date was continued until at least 20 larvae were found by digging the roots with a spatula. As soon as an immature planthopper was found, the depth of its position was measured with a ruler. The mean depth was calculated from the data of the planthoppers that were found at a particular date.

Results and discussion

The parameters that led to a minimal variation of the temperature sums from different years are listed in table 2. Most homogenous data could be achieved with parameter sets 5 and 6, respectively. However, all the data sets where the fourth day of the trap exposure period was

Table 2. Optimal parameters (start date and threshold temperature) for the prediction of flight of *Hyalesthes obsoletus*. Flight is expected when the mean sum of day-degrees is reached. AT=Daily average of air temperature; ST=Daily minimum of temperature on the soils surface.

Selected parameter set		Parameters for calculation of temperature sums						Difference between observed and calculated starting dates [days][1]			
		Day of the one week trap exposure period considered as start of flight	Start Date	Threshold temperature [°C]	Day-degrees till beginning of flight			Compared to sticky trap catches		Compared to life catches	
					Mean	SD	RSE [%]	Mean	SD	Mean	SD
1	AT optimal precision	1	March 9	5.8	1053	68	1.95	-0.8	2.8	2.2	5.1
2	AT convenient date	1	March 16	5.8	1008	73	2.19	-1.1	2.6	1.3	5.1
3	ST optimal precision	1	April 18	2.6	415	29	2.18	0.3	2.8	2.2	4.8
4	ST convenient date	1	Apr 01	2.6	465	34	2.32	0.5	3.6	2.2	5.7
5	AT optimal prec.+date	4	March 16	6.8	1021	38	1.18	0.7	2.9	-0,4	3.6
6	ST optimal precision	4	April 18	3.7	442	26	1.87	-0.7	3.2	-3.1	6.6
7	ST convenient date	4	Apr 01	2.6	499	30	1.90	-0.8	3.3	-3.1	6.7

[1] negative values represent a delayed, positive values a premature prognosis

Table 3. Comparison of observed and predicted flight data of adult *H. obsoletus*. The first day of sticky trap exposure was considered to be the observed start of flight activity for comparison with the predicted dates.

Year	Observed data			Prognosis based on parameter set 1			Prognosis based on parameter set 3		
	First sticky trap catches		First life catches	Date	Days difference to observed data[1]		Date	Days difference to observed data[1]	
	[1st day of exposure]	[4th day of exposure]			Sticky trap	Life catches		Sticky trap	Life catches
1993	01. Jun	04. Jun		01. Jun	0		01. Jun	0	
1994	16. Jun	19. Jun		13. Jun	3		10. Jun	6	
1995	12. Jun	15. Jun		16. Jun	-4		10. Jun	2	
1996	11. Jun	14. Jun		13. Jun	-2		11. Jun	0	
1999	08. Jun	11. Jun	15. Jun	03. Jun	5	12	06. Jun	2	9
2000	29. Mai	01. Jun	04. Jun	01. Jun	-3	3	28. Mai	1	7
2001	06. Jun	09. Jun	07. Jun	07. Jun	-1	0	08. Jun	-2	-1
2002	03. Jun	06. Jun	03. Jun	05. Jun	-2	-2	05. Jun	-2	-2
2003	02. Jun	05. Jun	04. Jun	03. Jun	-1	1	02. Jun	0	2
2004	07. Jun	10. Jun	09. Jun	10. Jun	-3	-1	11. Jun	-4	-2

[1]negative values represent a delayed, positive values a premature prognosis

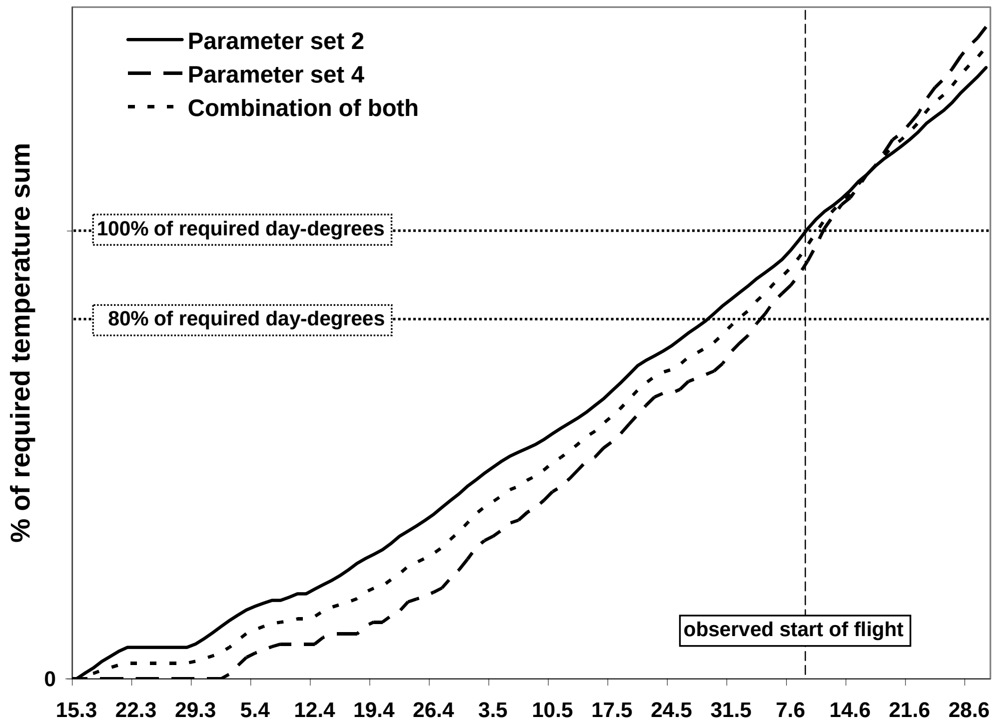

Fig. 1: Example from 2004 for the accumulation of temperature sums calculated with two of the parameter sets listed in table 1. Flight is expected as soon as 100 % of required day-degrees are reached. Traps should be exposed as soon as the day-degree sums get to 80 % of the other required values.

considered as start of the flight generally led to a delayed prediction, particularly when compared to the dates of life catches. Therefore, the parameter sets 1 to 4 appeared to be more suitable for our purpose. RSE values of the more convenient data sets (start of data accumulation at March 16 [set 2] and April 1 [set 4], respectively) were a little higher compared to the optimal sets 1 and 3. However, since the average differences of predicted dates between optimal and convenient parameter sets were less than one day, we decided to use parameter sets 2 and 4 for routine prediction. For practical use we combine the daily percentages of the required temperature sums (figure 1) calculated for both AT and ST. Exposure of sticky traps is recommended as soon as 80% of the required sums are reached. At the same time all weed control measures should be stopped in areas were the BN phytoplasma is present in order to avoid forced migration of emerging adults to grapevine which could increase the infection pressure. Life catches are most efficient during approximately four weeks after the start of the flight.

The beginning of flight activity of *H. obsoletus* as determined by sticky trap catches varied from May 29 in 2000 to June 16 in 1994 (table 3) with an average starting date of June 6 ± 5.6 days when the first day of the trap exposure period was considered as the start of the flight. With the fourth day as assumed starting date, the average was June 9 ± 5.6 days. First insects were caught alive between June 2 in 2000 and June 15 in 1999 (average and SD: June 7 ± 4.5 days). The flight lasted an average of 53 ± 5.7 days (from 47 to 60 days).

The method presented here allows the determination of appropriate day-degree sums for the prediction of the flight of *H. obsoletus*, although thermal thresholds and temperature sums for the development of this planthopper are not known because it cannot be reared under controlled conditions. By the variation of starting dates and threshold temperatures for accumulating temperature sums it was possible to identify combinations of these parameters that resulted in a minimum deviation of observed and predicted flight data and allowed the prediction of flight of the vector with sufficient reliability.

Table 4. Depth of the soil where larval instars of *H. obsoletus* were found on roots of *Ranunculus* sp. during winter of 2003/2004.

Date	Larval instars of H. obsoletus		
	n	Depth [cm]	
		Mean	SD
Oct 29	24	11.6	2.4
Nov 11	25	11.5	1.7
Dec 18	25	18.0	4.9
Jan 27	22	14.8	2.5
Feb 11	23	16.0	4.2
Mar 24	25	15.6	3.5
Mai 05	12	9.0	1.3

It is not clear what triggers the downward and upward movement of larvae in the soil. We found them at a maximum depth of 18 ± 4.9 cm in December (table 4), 9 ± 1.3 cm in the beginning of May and just below the soil surface under gravel later on. It can be assumed that air or surface temperatures do not become effective before the insects are rather close to the soil surface. This is likely to be the biological significance of the determined starting date for the accumulation of temperature sums. Since temperature effects might be interacting with intrinsic biological characteristics of the insects that have an effect on its movement in the soil and probably also with additional factors, e.g. soil structure or root system of the host plant, it appears to be necessary to check the suitability of the parameters determined in this work for different regions. It could also be possible, that temperature affects *H. obsoletus* only indirectly through the phenology of its host plants. Furthermore, differences between populations of *H. obsoletus* on nettle and bindweed have been observed. For example, the start of flight of planthoppers developing on nettle is delayed by approximately three weeks compared to bindweed (Langer & Maixner, unpublished). The method is therefore limited to populations on bindweed so far. More flight data from the planthopper populations on nettle are required before the method can be adjusted to this system. As soon as those data are available, it should be possible to predict the flight of *H. obsoletus* wherever appropriate weather data are available.

The procedure presented here allows the determination of relevant parameters for the prediction of insect development based on temperature sums even if the basic biological data cannot be defined in laboratory studies. It might be useful for other insects beside *H. obsoletus*, too.

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