

Modeling of the Bacterial Growth Curve

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Several sigmoidal functions (logistic, Gompertz, Richards, Schnute, and Stannard) were compared to describe a bacterial growth curve. They were compared statistically by using the model of Schnute, which is a comprehensive model, encompassing all other models. The t test and the F test were used. With the t test, confidence intervals for parameters can be calculated and can be used to distinguish between models. In the F test, the lack of fit of the models is compared with the measuring error. Moreover, the models were compared with respect to their ease of use. All sigmoidal functions were modified so that they contained biologically relevant parameters. The models of Richards, Schnute, and Stannard appeared to be basically the same equation. In the cases tested, the modified Gompertz equation was statistically sufficient to describe the growth data of *Lactobacillus plantarum* and was easy to use.

Predictive modeling is a promising field of food microbiology. Models are used to describe the behavior of microorganisms under different physical or chemical conditions such as temperature, pH, and water activity. These models allow the prediction of microbial safety or shelf life of products, the detection of critical parts of the production and distribution process, and the optimization of production and distribution chains. In order to build these models, growth has to be measured and modeled. Bacterial growth often shows a phase in which the specific growth rate starts at a value of zero and then accelerates to a maximal value (μ_m) in a certain period of time, resulting in a lag time (λ). In addition, growth curves contain a final phase in which the rate decreases and finally reaches zero, so that an asymptote (A) is reached. When the growth curve is defined as the logarithm of the number of organisms plotted against time, these growth rate changes result in a sigmoidal curve (Fig. 1), with a lag phase just after $t = 0$ followed by an exponential phase and then by a stationary phase.

Growth curves are found in a wide range of disciplines, such as fishery research, crop science, and biology. Most living matter grows with successive lag, growth, and asymptotic phases; examples of quantities that follow such growth curves are the length or mass of a human, a potato, or a fish and the extent of a population of fish or microorganisms. In addition, these sigmoidal curves are used in medical science for dose-mortality relations.

To describe such a curve and to reduce measured data to a limited number of interesting parameters, investigators need adequate models. A number of growth models are found in the literature, such as the models of Gompertz (7), Richards (14), Stannard et al. (17), Schnute (16), and the logistic model and others (15). These models describe only the number of organisms and do not include the consumption of substrate as a model based on the Monod equation would do. The substrate level is not of interest in our application, as we assume that there is sufficient substrate to reach intolerable numbers of organisms.

Besides the lag period and the asymptotic value, another valuable parameter of the growth curve is the maximum specific growth rate (μ_m). Since the logarithm of the number

is used, μ_m is given by the slope of the line when the organisms grow exponentially. Usually this parameter is estimated by deciding subjectively which part of the curve is approximately linear and then determining the slope of this curve section, eventually by linear regression (Table 1). A better method is to describe the entire set of data with a growth model and then estimate μ_m , λ , and A from the model. Some authors indeed use growth models to describe their data (Table 1). These models describe the number of organisms (N) or the logarithm of the number of organisms [$\log(N)$] as a function of time.

The motivation for the decision to use a given model is usually not stated. Only Gibson et al. (5) found better results by a fitting procedure with the Gompertz model when they compared that model with the logistic model. A large number of models as given in Table 1 are used, all more or less complicated and with different numbers of parameters. It can be expected that a difference in the results of the models for our application exists. Besides, the models are not written in terms of growth rate, lag time, and asymptotic value, which makes interpretation of the parameter values difficult.

The objective of this work is to evaluate similarities and differences between the models and to deal with the question of which model(s) can be used, on the basis of statistical reasoning. The models are rewritten in such a way that they contain parameters that are microbiologically relevant.

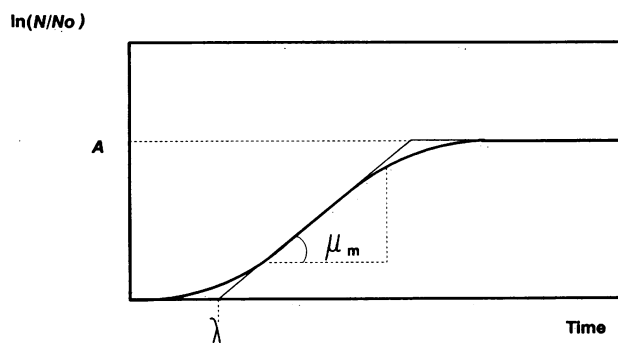


FIG. 1. A growth curve.

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TABLE 1. Some growth models used in the literature

Author(s)	Modeling	Model(s)
Adair et al. (1)	log(<i>N</i>)	Linear regression
Bratchell et al. (2)	log(<i>N</i>)	Gompertz
Broughall et al. (3)	log(<i>N</i>)	Linear regression
Einarsson and Eriksson (4)	log(<i>N</i>)	Logistic, polynomial
Gibson et al. (5)	log(<i>N</i>)	Logistic, Gompertz
Gibson et al. (6)	log(<i>N</i>)	Gompertz
Griffiths and Phillips (8)	log(<i>N</i>)?	Stannard
Jason (9)	<i>N</i>	Logistic
Mackey and Kerridge (10)	<i>N</i>	Gompertz
Phillips and Griffiths (12)	log(<i>N</i>)?	Stannard
Stannard et al. (17)	log(<i>N</i>)?	Stannard

THEORY

Description of the bacterial growth curve. Since bacteria grow exponentially, it is often useful to plot the logarithm of the relative population size [$y = \ln(N/N_0)$] against time (Fig. 1). The three phases of the growth curve can be described by three parameters: the maximum specific growth rate, μ_m , is defined as the tangent in the inflection point; the lag time, λ , is defined as the *x*-axis intercept of this tangent; and the asymptote [$A = \ln(N_\infty/N_0)$] is the maximal value reached. Curves may show a decline. This kind of behavior is called the death phase and is not considered in this paper.

Reparameterization of the growth models. Most of the equations describing a sigmoidal growth curve contain mathematical parameters (a , b , c , . . .) rather than parameters with a biological meaning (A , μ_m , and λ). It is difficult to estimate start values for the parameters if they have no biological meaning. Moreover, it is difficult to calculate the 95% confidence intervals for the biological parameters if they are not estimated directly in the equation but have to be calculated from the mathematical parameters. Therefore, all the growth models were rewritten to substitute the mathematical parameters with A , μ_m , and λ . This was done by deriving an expression of the biological parameters as a function of the parameters of the basic function and then substituting them in the formula. As an example, we show here the modification of the Gompertz equation, which is written as:

$$y = a \cdot \exp[-\exp(b - ct)] \quad (1)$$

To obtain the inflection point of the curve, the second derivative of the function with respect to t is calculated:

$$\frac{dy}{dt} = ac \cdot \exp[-\exp(b - ct)] \cdot \exp(b - ct) \quad (2)$$

$$\frac{d^2y}{dt^2} = ac^2 \exp[-\exp(b - ct)] \cdot \exp(b - ct) \cdot [\exp(b - ct) - 1] \quad (3)$$

At the inflection point, where $t = t_i$, the second derivative is equal to zero:

$$\frac{d^2y}{dt^2} = 0 \rightarrow t_i = b/c \quad (4)$$

Now an expression for the maximum specific growth rate can be derived by calculating the first derivative at the inflection point.

$$\mu_m = \left(\frac{dy}{dt} \right)_{t_i} = \frac{ac}{e} \quad (5)$$

The parameter c in the Gompertz equation can be substituted for by $c = \mu_m e/a$.

The description of the tangent line through the inflection point is:

$$y = \mu_m \cdot t + \frac{a}{e} - \mu_m t_i \quad (6)$$

The lag time is defined as the t -axis intercept of the tangent through the inflection point:

$$0 = \mu_m \cdot \lambda + \frac{a}{e} - \mu_m t_i \quad (7)$$

Using equations 4, 5, and 7 yields:

$$\lambda = \frac{(b - 1)}{c} \quad (8)$$

The parameter b in the Gompertz equation can be substituted by:

TABLE 2. Models used and their modified forms

Model	Equation	Modified equation ^a
Logistic	$y = \frac{a}{1 + \exp(b - cx)}$	$y = \frac{A}{1 + \exp\left[\frac{4\mu_m}{A}(\lambda - t) + 2\right]}$
Gompertz	$y = a \cdot \exp[-\exp(b - cx)]$	$y = A \exp\left\{-\exp\left[\frac{\mu_m \cdot e}{A}(\lambda - t) + 1\right]\right\}$
Richards	$y = a \{1 + v \cdot \exp[k(\tau - x)]\}^{(-1/v)}$	$y = A \left\{1 + v \cdot \exp(1 + v) \cdot \exp\left[\frac{\mu_m}{A} \cdot (1 + v) \left(1 + \frac{1}{v}\right) \cdot (\lambda - t)\right]\right\}^{(-1/v)}$
Stannard	$y = a \left\{1 + \exp\left[-\frac{(l + kx)}{p}\right]\right\}^{(-p)}$	$y = A \left\{1 + v \cdot \exp(1 + v) \cdot \exp\left[\frac{\mu_m}{A} \cdot (1 + v) \left(1 + \frac{1}{v}\right) \cdot (\lambda - t)\right]\right\}^{(-1/v)}$
Schnute	$y = \left\{y_1^b + \left(y_2^b - y_1^b\right) \cdot \frac{1 - \exp[-a(t - \tau_1)]}{1 - \exp[-a(\tau_2 - \tau_1)]}\right\}^{1/b}$	$y = \left(\frac{\mu_m (1 - b)}{a}\right) \left[\frac{1 - b \cdot \exp(a \cdot \lambda + 1 - b - at)}{1 - b}\right]^{1/b}$

^a $e = \exp(1)$.

TABLE 3. Selection of models based on Schnute (16)

Values of a and b	Model	No. of parameters
$a > 0, b = 0$	Gompertz	3
$a > 0, b < 0$	Richards	4
$a > 0, b = -1$	Logistic	3
$a = 0, b = 1$	Linear	2
$a = 0, b = 0.5$	Quadratic	2
$a = 0, b = 0$	t th power	2
$a < 0, b = 1$	Exponential	3

$$b = \frac{\mu_m e}{a} \lambda + 1 \quad (9)$$

The asymptotic value is reached for t approaching infinity:

$$t \rightarrow \infty: y \rightarrow a \Rightarrow A = a \quad (10)$$

The parameter a in the Gompertz equation can be substituted for by A , yielding the modified Gompertz equation:

$$y = A \exp \left\{ - \exp \left[\frac{\mu_m e}{A} (\lambda - t) + 1 \right] \right\} \quad (11)$$

The models with four parameters also contain a shape parameter (ν).

Table 2 shows the results for all equations used in this article.

The modified Stannard equation appears to be the same as the modified Richards equation. The parameters a and b in the Schnute equation are retained in the modified Schnute equation because they may be used for model selection (Table 3). However, substitution of a and b in the Schnute equation would result in the modified Richards equation.

Broughall et al. (3) used the Verhulst differential equation (resulting in a logistic curve) at times greater than the lag time and used $N = N_0$ if the time was smaller than the lag time. This relation has no smooth transition from a lag phase to a growth phase. Since all our growth data show such a smooth transition, this model was not considered.

Fitting of the data. The nonlinear equations were fitted to growth data by nonlinear regression with a Marquardt algorithm (11, 13). This is a search method to minimize the sum of the squares of the differences between the predicted and measured values. The program automatically calculates starting values by searching for the steepest ascent of the curve between four datum points (estimation of μ_m), by intersecting this line with the x axis (estimation of λ), and by taking the final datum point as estimation for the asymptote (A). The algorithm then calculates the set of parameters with the lowest residual sum of squares (RSS) and their 95% confidence intervals.

Model comparison. One way to discriminate among models is to compare them statistically. In that case, the RSS alone does not give enough information because different models can have a different number of parameters. Models with a greater number of parameters usually give a lower RSS. A better method is to determine whether it is worthwhile to use more parameters to lower the RSS. Therefore, data fits obtained by using the various models were compared statistically by the use of the t test and the F ratio test.

t test. First, the data were fitted by the Schnute model and parameters a and b were evaluated. The Schnute model is a comprehensive model; it encompasses all of the other simpler models. This is shown in Table 3, in which values of the

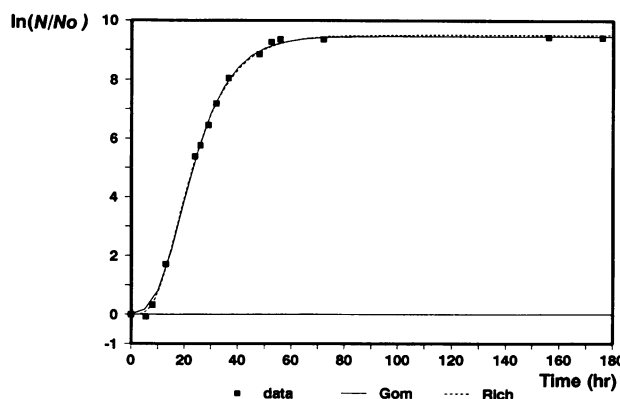


FIG. 2. Growth curve of *L. plantarum* at 18.2°C fitted with the Gompertz (Gom) and Richards (Rich) models.

Schnute parameters a and b are given, leading to one of the other models. The 95% confidence intervals of the different parameters were calculated with the value given by the Student t test. If, for instance, a value of zero is in the 95% confidence interval of b (and $a > 0$), the Gompertz model is suitable (Table 3).

F test. The logistic, Gompertz, Richards, and Schnute models were used to fit the data, and the RSS was calculated. Under the assumption that the four-parameter Schnute model exactly predicts the number of organisms, the RSS of the Schnute model was taken as an estimate of the measuring error. Whether a three-parameter model would be sufficient to describe the data could then be validated with an F test. In this test, the difference between the RSS values for the three- and four-parameter models was compared to the RSS of the four-parameter model. The difference in RSS of the three- and the four-parameter models is the profit we get from adding one parameter. If this profit is much smaller than the measuring error, as determined from the four-parameter model, adding the extra parameter is not worthwhile, as it would not be observable. If, however, this profit is much greater than the measuring error, it is worthwhile to add the extra parameter. The following is then calculated:

$$f = \frac{(RSS_2 - RSS_1)/(DF_2 - DF_1)}{RSS_1/DF_1} \text{ tested against } F_{DF_2 - DF_1}^{DF_1}$$

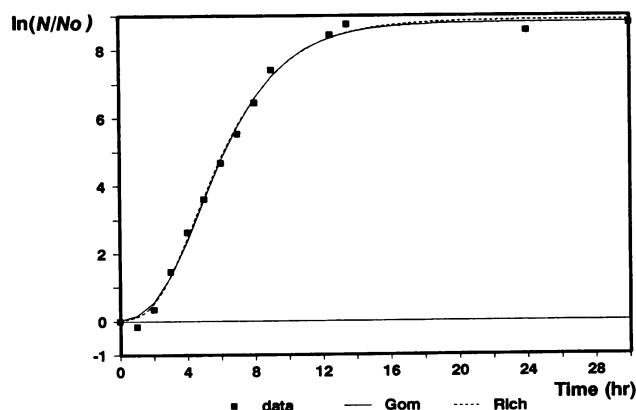


FIG. 3. Growth curve of *L. plantarum* at 35.0°C fitted with the Gompertz (Gom) and Richards (Rich) models.

TABLE 4. Statistical-analytical data for 40 growth curves of *L. plantarum*

Temp (°C)	$a_{\min}^{a,b}$	$b_{\min}^{a,b,c}$	b_{\max}	f^d		F table	RSS		
				Gompertz	Logistic		Gompertz	Logistic ^e	Richards
6.0	0.003	-0.988	0.994	0.001	0.078	4.13	1.70	1.70	1.70
6.1	-0.0006	-1.18	1.98	0.194	1.49	4.75	0.282	0.312	0.277
8.3	0.011	-0.113	0.435	1.91	53.6	4.21	0.707	1.97	0.660
8.6	0.008	-0.748	0.470	0.204	4.21	4.33	1.09	1.30	1.08
12.0	0.024	-0.247	0.548	0.577	18.4	4.36	1.13	2.11	1.10
12.2	0.023	-0.337	0.672	4.10	40.6	4.97	0.424	1.56	0.300
15.1	0.049	-0.786	0.289	0.875	5.03	4.45	1.06	1.30	1.00
15.2	0.049	-0.096	0.501	1.80	46.2	4.84	0.375	1.67	0.322
18.0	0.065	-0.086	0.540	7.84	49.9	4.60	0.611	2.02	0.391
18.2	0.059	-4.23	0.275	1.45	0.485	4.75	1.39	1.29	1.24
18.2	0.076	-0.058	0.471	3.78	60.4	4.84	0.233	1.12	0.173
18.2	0.068	-0.192	0.350	0.301	30.0	4.31	0.516	1.20	0.509
18.6	-0.607	-4.72	6.39	1.99	6.35	6.60	0.155	0.252	0.111
21.5	-0.322	-2.04	2.89	0.277	2.50	5.99	0.486	0.658	0.464
21.5	0.092	-0.167	0.614	1.05	19.2	4.75	0.648	1.55	0.596
25.0	0.116	-0.317	0.786	3.10	23.8	4.97	0.800	2.07	0.611
25.0	0.082	-0.147	0.970	2.44	16.8	4.84	1.07	2.21	0.876
28.4	0.259	-0.676	0.201	1.69	10.0	4.84	0.430	0.714	0.373
28.6	0.252	-0.352	0.183	0.481	34.5	4.84	0.210	0.833	0.201
32.0	0.257	-1.06	0.432	1.06	2.71	4.97	0.883	1.01	0.798
32.0	0.260	-0.933	0.367	1.33	4.85	5.59	0.334	0.475	0.281
32.4	0.272	-0.276	0.328	0.035	32.4	4.84	0.241	0.948	0.240
34.9	0.301	-0.544	0.421	0.099	114	5.59	0.200	3.41	0.197
35.0	0.255	-0.314	0.623	0.510	17.0	4.97	0.482	1.24	0.459
35.3	0.257	-0.320	0.455	0.136	21.1	4.75	0.471	1.28	0.466
36.6	0.262	-0.207	0.635	1.08	22.2	4.67	0.722	1.81	0.667
37.9	0.245	-0.299	0.676	0.604	15.4	4.60	0.880	1.77	0.844
38.4	0.264	-0.870	0.487	0.469	5.11	5.99	0.229	0.394	0.213
40.0	0.247	-1.73	0.391	2.65	0.382	4.84	1.17	0.974	0.941
41.4	0.151	-1.26	0.713	0.416	1.65	5.12	0.668	0.756	0.638
41.5	0.043	-0.220	0.855	5.18	29.1	4.75	0.331	0.792	0.231
41.5	0.070	-0.403	1.26	1.06	6.45	4.75	1.57	2.22	1.44
41.8	-0.278	-0.641	1.69	0.899	4.76	6.60	0.209	0.346	0.178
41.9	-1.01	-4.50	5.40	0.464	1.49	7.72	0.844	1.04	0.757
42.1	0.156	-0.152	0.726	3.31	26.8	4.67	0.400	0.975	0.319
42.2	0.014	-0.355	1.46	1.93	6.15	4.67	1.47	1.89	1.28
42.6	-0.101	-0.443	2.44	3.20	6.11	6.60	0.633	0.858	0.386
42.8	-7.82	-52.1	46.0	1.79	0.395	5.12	0.633	0.551	0.528
42.8	0.315	-1.09	0.824	0.083	2.63	4.75	0.141	0.171	0.140
42.8	-0.061	-2.20	1.61	0.084	0.527	5.12	0.105	0.110	0.104

^a a and b are Schnute parameters.^b min and max are 95% confidence limits.^c Boldface data indicate acceptance of logistic model with t test.^d Boldface data indicate acceptance of given model with F test.^e Boldface data indicate that RSS with Gompertz model is greater than RSS with logistic model.

where RSS_1 is the RSS from the Schnute model, RSS_2 is the RSS from the three-parameter model, DF_1 is the number of degrees of freedom from the Schnute model and equals n points - 4, and DF_2 is the number of degrees of freedom from the three-parameter model and equals n points - 3. Note that $DF_2 - DF_1 = 1$, so the F test becomes:

$$f = \frac{RSS_2 - RSS_1}{RSS_1/DF_1} \text{ tested against } F_{DF_1}^1$$

If the models were linear in their parameters, this f value would be F -distributed under the assumption that the four-parameter model is correct. Even for nonlinear models, the variance ratio shown above is approximately F -distributed when the sample size is large (16). This analysis is an approximation at best, and this procedure should be considered an informal process, rather than a rigorous statistical analysis, because of the use of nonlinear models (16). In

TABLE 5. Determination of models for *L. plantarum* based on the method of Schnute (16)

Values of a and b	Model	No. (% of total) ^a of results accepted with:	
		t test	F test
$a > 0, b = 0$	Gompertz	40 (100)	38 (95)
$a > 0, b < 0$	Richards	40 (100)	
$a > 0, b = -1$	Logistic	11 (28)	17 (43)
$a = 0, b = 1$	Linear	8 (20)	
$a = 0, b = 0.5$	Quadratic	8 (20)	
$a = 0, b = 0$	r th power	8 (20)	
$a < 0, b = 1$	Exponential	8 (20)	

^a Total number of experiments = 40.

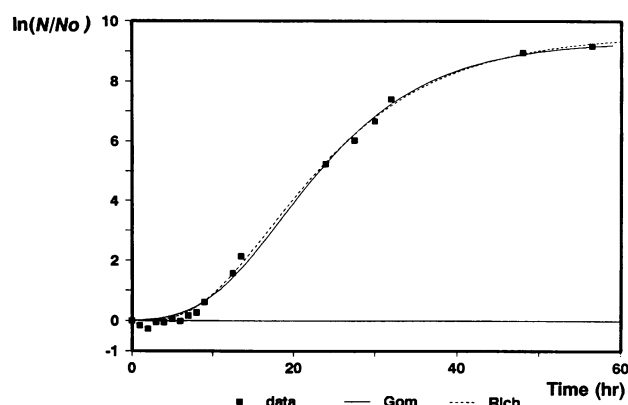


FIG. 4. Growth curve of *L. plantarum* at 18.0°C fitted with the Gompertz (Gom) and Richards (Rich) models.

some boundary cases, the Student *t* test and the *F* test can therefore give contradictory results.

MATERIALS AND METHODS

In 40 experiments, *Lactobacillus plantarum* (American Type Culture Collection [ATCC]-identified; no ATCC number) was cultivated in MRS medium (Difco Laboratories) at different temperatures. Growth was measured with plate counts on pour plates (MRS medium with 12 g of agar [Agar technical; Oxoid Ltd.] per liter). The inoculation level was 0.01% (about $5 \cdot 10^5$ organisms).

Growth data of *Candida parapsilosis*, *Pseudomonas putida*, *Enterobacter agglomerans*, a *Nocardia* sp., *Salmonella heidelberg*, *Staphylococcus aureus*, and *Listeria monocytogenes* were kindly provided by J. P. M. Smelt, C. J. M. Winkelmolen, P. Breeuwer, and F. G. C. T. Sommerdijk.

RESULTS

All of the models visually gave reasonably good fits of the data (Fig. 2 and 3, for example). In some cases, the Schnute and Richards models gave some problems with the fitting because the parameter estimates came in an area where the function predicted such a large value that an overflow error resulted. The Gompertz and logistic models never gave

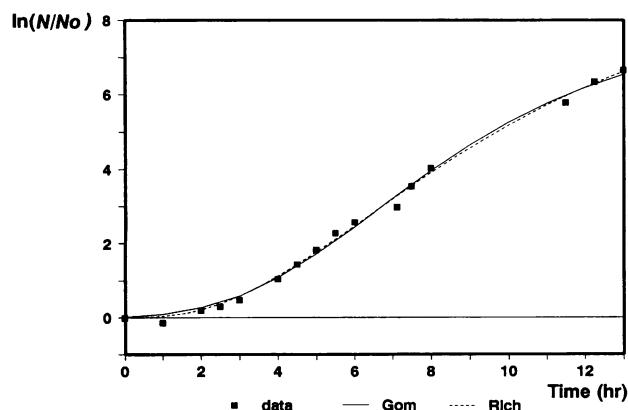


FIG. 5. Growth curve of *L. plantarum* at 41.5°C fitted with the Gompertz (Gom) and Richards (Rich) models.

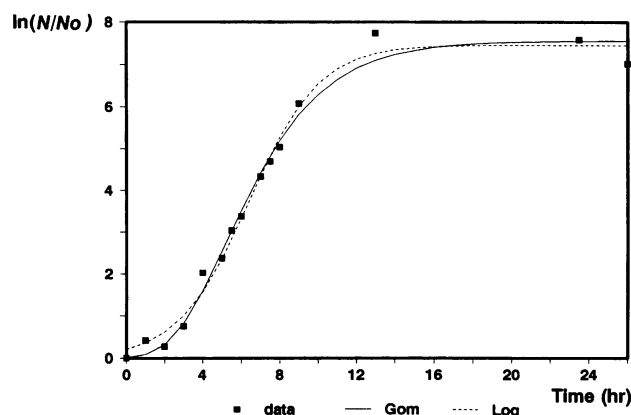


FIG. 6. Growth curve of *L. plantarum* at 40.0°C fitted with the Gompertz (Gom) and logistic (Log) models.

problems with fitting. In all cases, the RSS values for the Richards and Schnute models were the same, which was expected because the models are basically the same.

Plate count data for *L. plantarum*. In Table 4, the results of the parameter estimation for 40 sets of data are reported. In this table, the temperature at which the experiment was conducted is given. With the use of the Student *t* test value, the 95% confidence intervals for the parameters *a* and *b* were calculated. The lower 95% confidence limit of the parameter *a* is given in Table 4 to determine whether *a* = 0 is within the confidence interval. Furthermore, the 95% confidence limits for *b* are given. Comparing the confidence intervals in Table 4 for *b* with Table 3 results in Table 5. In Table 3, we can see that if *b* = 0, the Schnute model changes into the Gompertz model, so we accepted the Gompertz model if the value of zero was within the 95% confidence interval of *b*. This was true in all cases (Table 4), so the Gompertz model, although a three-parameter model, was accepted in all cases by the *t* test. Furthermore, the *f*-testing values for the Gompertz and the logistic models and the *F* table values are given in Table 4. With the use of the *F* test, the difference between the RSS values for the three- and four-parameter models was compared to the RSS of the four-parameter model. For the Gompertz model, the *f*-testing value was lower than the *F* table value in all but two cases (Table 4).

In the cases in which the *F* test favored the Richards

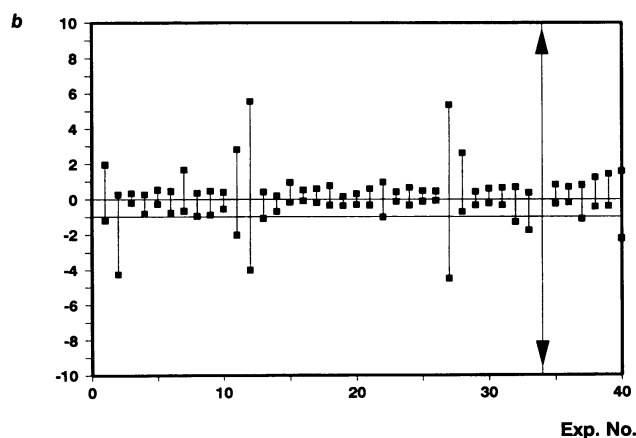


FIG. 7. *b* confidence intervals of *L. plantarum* growth data fitted with the Schnute model (16). Exp. No., Experiment number.

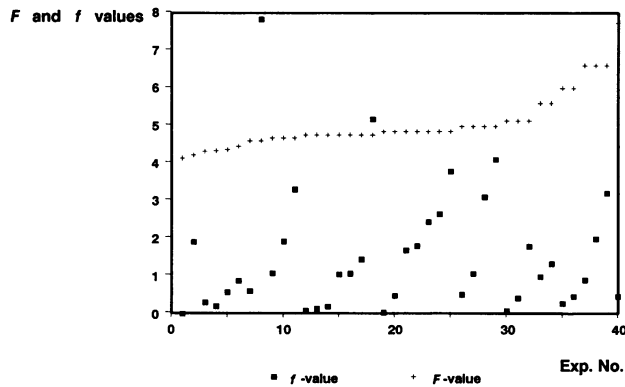


FIG. 8. Results of an F test of *L. plantarum* growth data. The Gompertz and Richards models are compared.

model over the Gompertz model (Fig. 4 and 5), the differences between the two models were still very small. The logistic model, however, was accepted by the t test only 11 times (out of 40) and by the F test 17 times (Table 4).

In addition, the RSS values of the Gompertz, logistic, and Richards models are given in Table 4. The RSS values for the four-parameter models were always lower than the RSS values for the three-parameter models. In only three cases, the logistic model gave a lower RSS value than the Gompertz model (Table 4; Fig. 6), but in these cases the Gompertz model still fitted the data acceptably.

In Fig. 7, the confidence intervals for the parameter b (Schnute model with the t test) are shown. In this graph, it can be seen that the value of zero (Table 3, Gompertz model)

was always within the confidence interval; however, the value of -1 (Table 3, logistic model) was much less frequently within the confidence interval (only 11 times).

In Fig. 8, the results from the F test for the Gompertz model are shown. The squares represent the f -testing values, and the pluses represent the critical F table values (95% confidence). If the f -testing value was smaller than the F table value, the three-parameter model was accepted. In this graph, it can be seen that the Gompertz model was rejected only 2 times out of 40 (5%). This 5% rejection level may be expected with a 95% confidence level.

Plate count data for other organisms. While it could be that only *L. plantarum* growth data are described well by the Gompertz model, the same comparison of models was carried out with growth data from other microorganisms (Tables 6 and 7). With these data, the Gompertz model was accepted in 70% of the cases by the t test ($b = 0$ within the confidence interval) and in 67% of the cases by the F test. The logistic model was accepted in 52% of the cases by the t test ($b = 0$ within the confidence interval) and in 59% of the cases by the F test.

DISCUSSION

In order to build models to describe the growth of microorganisms in food, it is necessary to measure growth curves. To reduce the measured data to interesting parameters such as the growth rate, it is recommended that the data be described with a model instead of by using linear regression over a subset of the data. Sigmoidal models to describe the growth data can be constructed with three or four parameters.

We compared several models statistically and found that,

TABLE 6. Statistical-analytical data for 27 growth curves of organisms other than *L. plantarum*

Organism	$a_{\min}^{a,b}$	$b_{\min}^{a,b,c}$	$b_{\max}^{b,c}$	f^d		F table	RSS		
				Gompertz	Logistic		Gompertz	Logistic ^e	Richards
<i>Candida parapsilosis</i>	0.038	-1.60	0.761	1.71	1.11	10.1	0.114	0.100	0.073
<i>C. parapsilosis</i>	0.136	-1.12	0.154	6.67	4.86	10.1	0.086	0.070	0.027
<i>C. parapsilosis</i>	0.039	-1.60	1.16	0.071	1.07	10.1	0.102	0.135	0.100
<i>C. parapsilosis</i>	0.117	-0.673	0.751	0.557	21.1	10.1	0.030	0.205	0.025
<i>C. parapsilosis</i>	-0.071	-3.68	1.70	3.22	0.000	10.1	0.362	0.175	0.175
<i>Pseudomonas putida</i>	0.050	-2.42	-0.513	18.7	1.08	4.17	4.47	2.86	2.76
<i>P. putida</i>	0.027	0.114	1.12	3.84	15.3	4.17	6.42	8.60	5.70
<i>P. putida</i>	0.059	-0.169	0.701	1.25	16.2	4.13	3.98	5.66	3.83
<i>P. putida</i>	0.037	-0.174	1.54	6.31	17.5	4.16	11.9	15.4	9.85
<i>Enterobacter agglomerans</i>	0.002	0.141	1.10	15.2	41.5	4.14	13.4	20.7	9.19
<i>E. agglomerans</i>	0.015	0.412	0.802	26.7	111	4.17	4.87	12.1	2.58
<i>E. agglomerans</i>	0.020	0.240	0.793	9.26	48.8	4.13	5.07	9.70	3.98
<i>E. agglomerans</i>	0.025	0.432	1.01	17.7	59.9	4.15	7.80	14.4	5.03
<i>Nocardia</i> sp.	0.072	-2.08	1.29	0.540	1.21	5.12	0.176	0.188	0.166
<i>Salmonella heidelberg</i>	-3.18	-10.9	12.9	0.720	1.43	161	1.23	1.74	0.717
<i>Staphylococcus aureus</i>	0.178	-0.759	1.82	0.843	3.86	6.61	0.526	0.798	0.450
<i>S. aureus</i>	0.009	-3.73	5.39	15.2	42.5	6.61	0.543	1.28	0.134
<i>S. aureus</i>	-2.13	-3.39	5.39	1.25	1.73	6.61	1.87	2.01	1.49
<i>S. aureus</i>	-3.60	-5.07	7.07	1.28	1.56	6.61	0.886	0.926	0.706
<i>S. aureus</i>	-0.529	-2.11	4.09	2.86	4.66	6.61	4.54	5.58	2.89
<i>S. aureus</i>	-0.315	-4.67	6.44	1.51	3.31	6.61	0.807	1.03	0.620
<i>S. aureus</i>	0.062	0.235	1.77	10.8	21.6	10.1	1.12	1.99	0.243
<i>S. aureus</i>	0.094	-0.056	2.06	14.4	29.6	10.1	0.873	1.63	0.151
<i>S. aureus</i>	0.452	0.747	1.25	3.44	5.31	10.1	0.273	0.353	0.127
<i>S. aureus</i>	0.066	-0.512	1.87	2.62	8.59	10.1	0.628	1.29	0.335
<i>S. aureus</i>	-16.5	-171	172	0.000	0.136	18.5	0.007	0.007	0.007
<i>Listeria monocytogenes</i>	-0.060	-1.66	1.81	0.094	3.20	18.5	0.031	0.078	0.030

^a a and b are Schnute parameters.

^b min and max are 95% confidence limits.

^c Boldface data indicate acceptance of logistic model with t test.

^d Boldface data indicate acceptance of given model with F test.

^e Boldface data indicate that RSS with Gompertz model is greater than RSS with logistic model.

TABLE 7. Determination of models for organisms other than *L. plantarum* based on the method of Schnute (16)

Values of a and b	Model	No. (% of total) ^a of results accepted with:	
		t test	F test
$a > 0, b = 0$	Gompertz	19 (70)	18 (67)
$a > 0, b < 0$	Richards	25 (93)	
$a > 0, b = -1$	Logistic	14 (52)	16 (59)
$a = 0, b = 1$	Linear	8 (30)	
$a = 0, b = 0.5$	Quadratic	8 (30)	
$a = 0, b = 0$	r th power	8 (30)	
$a < 0, b = 1$	Exponential	8 (30)	

^a Total number of experiments = 27.

for *L. plantarum*, the Gompertz model was accepted in all cases by the t test and was accepted in 95% of the cases by the F ratio test; therefore, the Gompertz model can be regarded as sufficient to describe the growth curves of *L. plantarum*. The logistic model, however, seems not to be sufficient to describe the data. It was accepted in 28% of the cases by the t test and in 43% of the cases by the F test with *L. plantarum*.

With the data of other microorganisms, the Gompertz model was accepted in 70% of the cases. With the data of the other organisms, the logistic model was accepted in 52% of the cases with the t test and in 59% of the cases with the F test. Linear, quadratic, r th-power, and exponential models were accepted in very few cases. Therefore, we can conclude that all growth curves are better fitted with the Gompertz model than with logistic, linear, quadratic, r th-power, and exponential models.

In some cases, the confidence interval of the Schnute parameter b ($b_{\min} - b_{\max}$) was very large. In these cases, there were not enough data to describe all three growth phases. Therefore, the confidence level of the resulting parameters is not very high. These sets of data are not very suitable for the estimation of parameters.

In a number of cases, the four-parameter Schnute model was statistically better than the Gompertz model (*P. putida* and *E. agglomerans*). These growth curves contained a very large number of datum points (34 to 38), and with such a large number of datum points the difference in degrees of freedom between three- and four-parameter models is not important (with 34 datum points: 31 degrees of freedom for Gompertz or 30 degrees of freedom for Richards). For the other organisms (*C. parapsilosis* and *S. aureus*), the Gompertz model was accepted in most cases. For the growth curves of the *Nocardia* sp., *Salmonella heidelberg*, and *Listeria monocytogenes*, the Gompertz model was accepted in all the cases, but only one curve for these organisms was used.

The three-parameter models gave no difficulties in finding the least-square parameters. In almost all the cases, the Gompertz model can be regarded as the best model to describe the growth data. If a three-parameter model is sufficient to describe the data, it is recommended over a four-parameter model because the three-parameter model is simpler and therefore easier to use and because the three-parameter solution is more stable since the parameters are less correlated. Moreover, when a three-parameter model is used, the estimates have more degrees of freedom, which can be important when a growth curve with a small number of measured points is used. Furthermore, it is very important that all three parameters can be given a biological meaning.

The fourth parameter in the four-parameter models is a shape parameter and is difficult to explain biologically.

In a number of cases (especially when a large number of datum points are collected), a four-parameter model can be significantly better; therefore, it is recommended that the procedure given in this paper be carried out with a number of sets of data in order to find out the best model to describe the specific sets of data.

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LITERATURE CITED

- Adair, C., D. C. Kilsby, and P. T. Whittall. 1989. Comparison of the Schoolfield (non-linear Arrhenius) model and the square root model for predicting bacterial growth in foods. *Food Microbiol.* 6:7-18.
- Bratchell, N., A. M. Gibson, M. Truman, T. M. Kelly, and T. A. Roberts. 1989. Predicting microbial growth: the consequences of quantity of data. *Int. J. Food Microbiol.* 8:47-58.
- Broughall, J. M., P. A. Anslow, and D. C. Kilsby. 1983. Hazard analysis applied to microbial growth in foods: development of mathematical models describing the effect of water activity. *J. Appl. Bacteriol.* 55:101-110.
- Einarsson, H., and S. G. Eriksson. 1986. Microbial growth models for prediction of shelf life of chilled meat, p. 397-402. *In* Recent advances and developments in the refrigeration of meat by chilling. Institut International du Froid-International Institute of Refrigeration, Paris.
- Gibson, A. M., N. Bratchell, and T. A. Roberts. 1987. The effect of sodium chloride and temperature on the rate and extent of growth of *Clostridium botulinum* type A in pasteurized pork slurry. *J. Appl. Bacteriol.* 62:479-490.
- Gibson, A. M., N. Bratchell, and T. A. Roberts. 1988. Predicting microbial growth: growth responses of salmonellae in a laboratory medium as affected by pH, sodium chloride and storage temperature. *Int. J. Food Microbiol.* 6:155-178.
- Gompertz, B. 1825. On the nature of the function expressive of the law of human mortality, and on a new mode of determining the value of life contingencies. *Philos. Trans. R. Soc. London* 115:513-585.
- Griffiths, M. W., and J. D. Phillips. 1988. Prediction of the shelf-life of pasteurized milk at different storage temperatures. *J. Appl. Bacteriol.* 65:269-278.
- Jason, A. C. 1983. A deterministic model for monophasic growth of batch cultures of bacteria. *Antonie van Leeuwenhoek J. Microbiol. Serol.* 49:513-536.
- Mackey, B. M., and A. L. Kerridge. 1988. The effect of incubation temperature and inoculum size on growth of salmonellae in minced beef. *Int. J. Food Microbiol.* 6:57-65.
- Marquardt, D. W. 1963. An algorithm for least-squares estimation of nonlinear parameters. *J. Soc. Ind. Appl. Math.* 11:431-441.
- Phillips, J. D., and M. W. Griffiths. 1987. The relation between temperature and growth of bacteria in dairy products. *Food Microbiol.* 4:173-185.
- Press, W. H., B. P. Flannery, S. A. Teukolsky, and W. T. Vetterling. 1988. *MRQMIN*, p. 772-776. *In* Numerical recipes. Cambridge University Press, Cambridge.
- Richards, F. J. 1959. A flexible growth function for empirical use. *J. Exp. Bot.* 10:290-300.
- Ricker, W. E. 1979. Growth rates and models. *Fish Physiol.* 8:677-743.
- Schnute, J. 1981. A versatile growth model with statistically stable parameters. *Can. J. Fish. Aquat. Sci.* 38:1128-1140.
- Stannard, C. J., A. P. Williams, and P. A. Gibbs. 1985. Temperature/growth relationship for psychrotrophic food-spoilage bacteria. *Food Microbiol.* 2:115-122.