Analysis of Progress Curves of Sugarcane Smut on Different Cultivars Using Functions of Double Sigmoid Pattern

L. Amorim, A. Bergamin Filho, and B. Hau

First and second authors: Depto. Fitopatologia, ESALQ-USP, Caixa Postal 9, 13400 Piracicaba, SP, Brazil; third author: Biometrie und Populationsgenetik, Justus-Liebig Universität, 6300 Giessen, Germany.

ABSTRACT

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The progress of smut on the sugarcane cultivars NA56-79, SP71-799, SP71-1406, SP71-6163, and SP70-1143 was quantified with artificial and natural inoculation during five consecutive years at Primeiro de Maio, Paraná, Brazil. The annual progress curves were fitted by the generalized monomolecular $y(t) = p_1 \{1 - \exp[-(p_2 + p_3 t + p_4 t^2 + p_5 t^3)]\}$ and the generalized Gompertz $y(t) = p_1 \exp\{-\exp[-(p_2 + p_3 t + p_4 t^2 + p_5 t^3)]\}$ functions, in which the dependent variable $y(t)$ was disease severity (whips per hectare) and the independent variable t was time (days after harvest).

Estimates of the upper asymptote, the mean infection rate, the transition time between growth waves of the annual disease progress curve, and the latent period, based on the generalized Gompertz function, were utilized to compare cultivars. None of the time-related curve elements, i.e., the transition time between growth waves and latent period, differentiated the cultivars because the values were similar for all cultivars. Cultivars NA56-79 and SP71-799 showed the highest and lowest upper asymptote, respectively, differing statistically ($P < 0.05$) from the others.

Ustilago scitaminea Syd. & P. Syd., the causal agent of sugarcane smut, produces one or two cycles of infection during each annual cycle of sugarcane production (2,10). Two cycles, expressed by the production of two distinct waves of whips (whiplike sori that arise either from the terminal meristem or from lateral shoots), are particularly evident in susceptible cultivars. The first cycle is due to primary infections while the second cycle is caused by secondary infections (1). These infection cycles lead to annual disease progress curves of double sigmoid pattern for the cumulative number of whips. Simple growth functions traditionally used in epidemiological studies, such as the logistic, Gompertz, Richards, and Weibull models, are not suitable for analysis of this type of disease progress curve because their shape is sigmoid. These models can be used to fit the annual epidemics of sugarcane smut only if the disease progress curves are subdivided into two parts that are analyzed separately. Amorim and Bergamin Filho (1) used this procedure to analyze the behavior of four sugarcane cultivars. In 60% of the 40 situations assessed by these authors, the first phase, or first wave, of the annual disease progress curve was best described by the monomolecular model. The second wave, in turn, was better fitted to the logistic model. Thus, two distinct infection rates, one for each wave, were obtained for each cultivar. Although this approach permits the characterization of cultivar resistance, it imposes some restrictions on the analysis of disease dynamics. For example, it is not possible to predict the progress of smut in the second cycle by knowing how it progressed in the first part of the epidemic. For the simulation of epidemics, the utilization of this methodology is not adequate because the transition moment between the two waves of the disease progress curve cannot be determined. In the cultivar analysis (1), the transition point of field data was defined as the moment when the least increase in the disease severity was found between two consecutive assessments.

Mathematical functions that are able to describe growth curves of double sigmoid pattern (7,9) seem to be promising in the study of the dynamics of this disease. Among the functions tested in previous studies (6), the generalizations of the monomolecular and Gompertz models were the most suitable to fit the increase in severity of sugarcane smut. With a single function capable

of fitting the entire disease progress curve, it is possible to go beyond the simple characterization of cultivar susceptibility and draw inferences about general aspects of the dynamics of the pathosystem, such as average rate of infection, duration of the latent period, asymptote for the first growth phase, transition time from the first to the second growth phase, and final asymptote.

MATERIAL AND METHODS

The disease progress curves analyzed in this study were obtained in an experiment conducted at the plant pathology farm of Copersucar's Technology Centre in Primeiro de Maio, Paraná, Brazil. The experiment, initiated in February 1985 and conducted through April 1990, included plant cane and four ratoon crops. ("Ratoon" refers to a shoot arising from a sugarcane crown after harvest, the first ratoon being the sprout after the first harvest). Five commercial sugarcane cultivars (NA56-79, SP71-799, SP71-1406, SP71-6163, and SP70-1143) were subjected to two treatments: a treatment with inoculation of *U. scitaminea* and a control treatment without inoculation. For the inoculation, cuttings (three-bud pieces) of sugarcane were dipped in a teliospore suspension (10^6 spores per milliliter) for 20 min before they were planted. The cuttings were distributed in a field with a randomized block design. There were three replicates. The experimental plot consisted of eight 10-m rows (with 20 three-bud cuttings of sugarcane per row) with spaces of 1.4 m between rows.

To assess disease progress, the cumulative number of apical whips in each plot were counted fortnightly or monthly. The new whips were marked with plastic tape in each assessment to differentiate them from the older ones.

No herbicides, insecticides, or fungicides were used during the course of the experiment so that there would be no interference with disease progress. Cultivation was done entirely with a hoe, and the field was irrigated by a sprinkler only once (immediately after the cuttings were planted). Fertilizer was applied at a rate of 500 kg/ha of 5-25-25 (N-P-K) at the time of planting or 18-00-27 immediately after the harvest of the ratoon crops.

Analysis of the data with functions of double sigmoid pattern. On the basis of results of previous studies (6), we chose two mathematical functions to fit the smut progress curves: the gen-

eralized monomolecular function with five parameters

$$y(t) = p_1 \{1 - \exp[-(p_2 + p_3 t + p_4 t^2 + p_5 t^3)]]\} \qquad (1)$$

and the generalized Gompertz function with five parameters

$$y(t) = p_1 \exp\{- \exp(-[p_2 + p_3 t + p_4 t^2 + p_5 t^3)]]\}, \qquad (2)$$

where the dependent variable $y(t)$ represents disease severity, assessed as whips per hectare, and the independent variable t represents time, measured in days after harvest. The polynomial combines initial disease level and infection rate parameters as a function of time in a nonlinear fashion.

For both functions, p_1 is the maximum disease level or upper asymptote, and p_2 is related to the initial disease level y_0 (y at $t = 0$) by $p_2 = \ln[p_1/(p_1 - y_0)]$ for the generalized monomolecular

function and $p_2 = -\ln[-\ln(y_0/p_1)]$ for the generalized Gompertz function. The parameters p_3, p_4, and p_5 describe the infection rate as a function of time ($r(t) = p_3 + 2p_4 t + 3p_5 t^2$) if the generalized functions are regarded as simple monomolecular or Gompertz functions (6).

For some of the disease progress curves, particularly those for the more resistant cultivars, two distinct growth waves were not detected. In these cases, the parameters p_4 and p_5 of the generalized functions were set to zero, and the infection rate remained constant, i.e., equal to p_3. Data were analyzed by nonlinear regression analysis, conducted by Marquardt's compromise method (4) with the software packages PlotIT (Scientific Programming Enterprises, Haslett, MI) (5) and BMDP (3).

Eight curve elements of the fitted generalized Gompertz function were used to compare cultivar response. The first three elements (times tp_1, tp_2, and tp_3) were estimated numerically from points of inflection on the progress curve (Fig. 1). At the time tp_1, when the first point of inflection occurs, the first wave of the derivative has a maximum value (Fig. 1). At tp_2, the transition point from the first to the second growth phase, the derivative takes its minimum value. The third point of inflection at time tp_3 coincides with the maximum value of the second wave of the derivative. The fourth element, t_{max}, is the time at which 99% of y_{max} is reached (Fig. 1). The other four elements of the progress curves are the disease levels at the three points of inflection, i.e., $y(tp_1)$, $y(tp_2)$, and $y(tp_3)$, and the maximum value y_{max} (Fig. 1), which is given by the parameter p_1 of the generalized Gompertz function (equation 2). In the nonlinear analysis, the latter parameter always showed low intercorrelation with other parameters of the equation, and it could be estimated with low standard error. Therefore, the parameter y_{max} was used to compare the progress curves of different cultivars statistically, using the confidence intervals for the curve asymptote.

The mean infection rate ($\bar{r}$) was determined by integration of the rate function $r(t)$ from the first (t_f) to the last (t_l) field
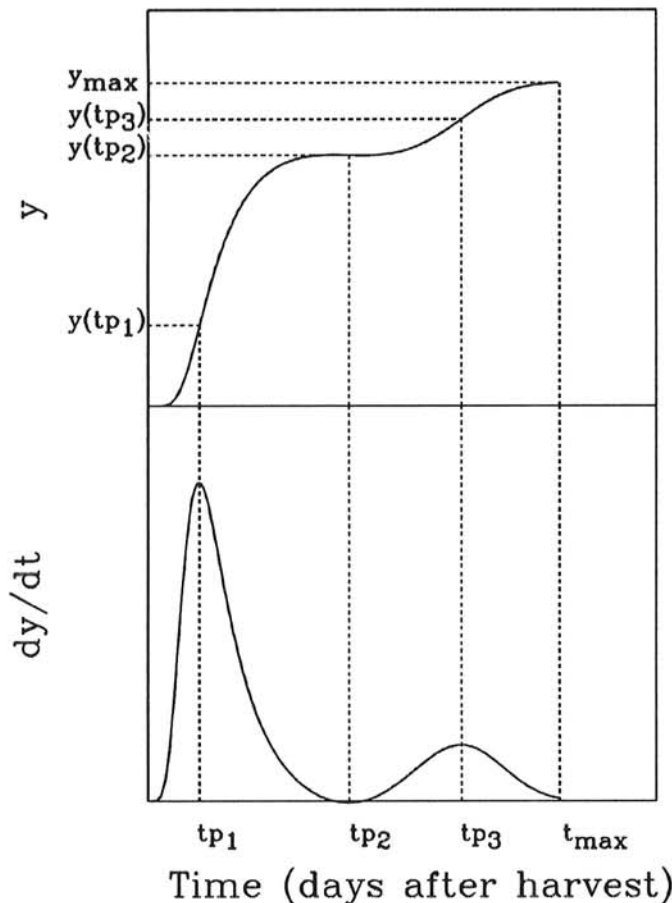

Fig. 1. Curve elements of the generalized Gompertz function used to compare cultivars. At times tp_1, tp_2, and tp_3 the progress curve has points of inflection, the extreme values of the derivative. Time t_{max} is defined as the point at which 99% of the upper asymptote (y_{max}) is reached. The corresponding disease levels are $y(tp_1)$, $y(tp_2)$, and $y(tp_3)$.

TABLE 1. Coefficients of determination (R^2) of the generalized monomolecular (GM) and generalized Gompertz (GG) functions fitted to the disease progress curves of sugarcane smut on ratoons of four commercial sugarcane cultivars dip-inoculated with *Ustilago scitaminea*

	Cultivar							
	NA56-79		SP71-1406		SP71-6163		SP71-799	
Ratoon	GM	GG	GM	GG	GM	GG	GM	GG
First	0.998	0.993	0.989	0.998	0.992	0.997	0.995*[a]	0.997*
Second	0.995	0.992	0.995	0.996	0.999	0.996	0.992*	0.998*
Third	0.992	0.985	0.997	0.992	0.988	0.974	0.992	0.976
Fourth	0.999	0.999	0.992	0.997	0.989	0.996	0.996	0.999

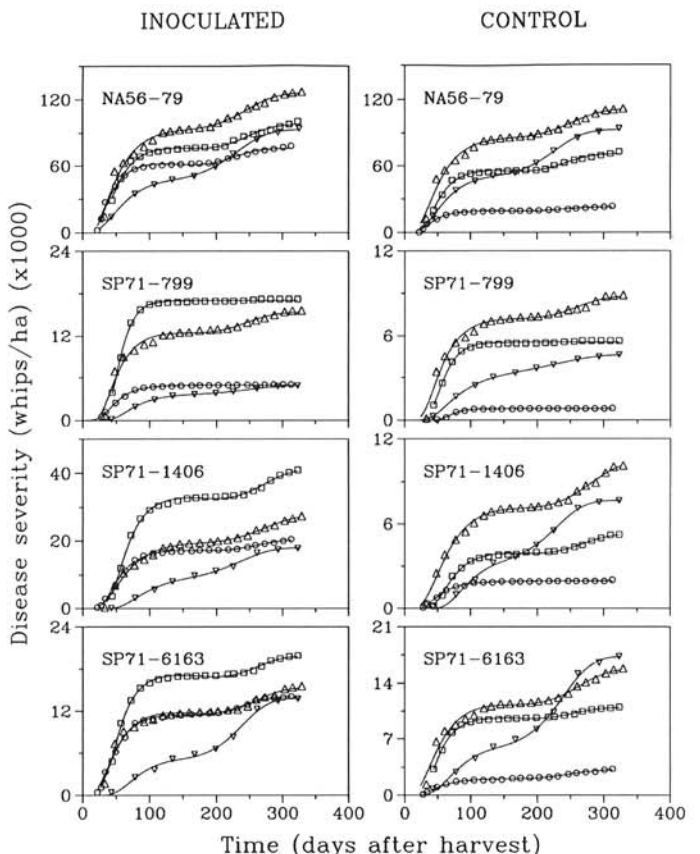[a] Asterisks indicate values obtained when p_4 and p_5 were equal to 0.

Fig. 2. Observed (symbols) and predicted (lines) disease progress curves of the severity of sugarcane smut in cultivars NA56-79, SP71-799, SP71-1406, and SP71-6163 in the first (○), second (□), third (△), and fourth (▽) ratoon stages obtained with the generalized Gompertz function.

observation measured in days. Therefore, the mean infection rate was computed as

$$\bar{r} = [p_3(t_1 - t_f) + p_4(t_1^2 - t_f^2) + p_5(t_1^3 - t_f^3)]/t_1 - t_f . \quad (3)$$

RESULTS

The plant-cane stage of all five cultivars and four ratoons of cultivar SP70-1143 had extremely low smut severity (1). Therefore, these disease progress curves were excluded from further analysis, and only the remaining 32 curves (four cultivars × two treatments × four ratoons) were fit with the generalized functions of monomolecular and Gompertz models that describe growth curves with double sigmoid pattern (equations 1 and 2).

Both functions provided excellent fit of data, as can be seen from the coefficients of determination in Table 1 for the inoculated treatment ($R^2 > 0.97$). Curves calculated with the generalized Gompertz function are illustrated for four cultivars and four ratoons in both treatments (Fig. 2).

The generalized Gompertz function was chosen to further analyze and compare progress curves of different cultivars because of its superior flexibility. The function fit data well even in cases where the second growth phase of the disease was inconspicuous. In addition, the generalized Gompertz function was used to estimate the latent period of the pathogen through time tp_1, when the derivative had a maximum value for the first wave (Fig. 1), which corresponded to the appearance of approximately 30% of the whips. Because the generalized monomolecular function had no point of inflection in the first growth phase, there was no local maximum that could be used to characterize the length of the latent period.

The derivative of the disease progress curves given by a generalized Gompertz function (equation 2) presented two waves for most cultivars (Fig. 3). The only exceptions occurred when disease progress curves did not have a second growth phase, for example,
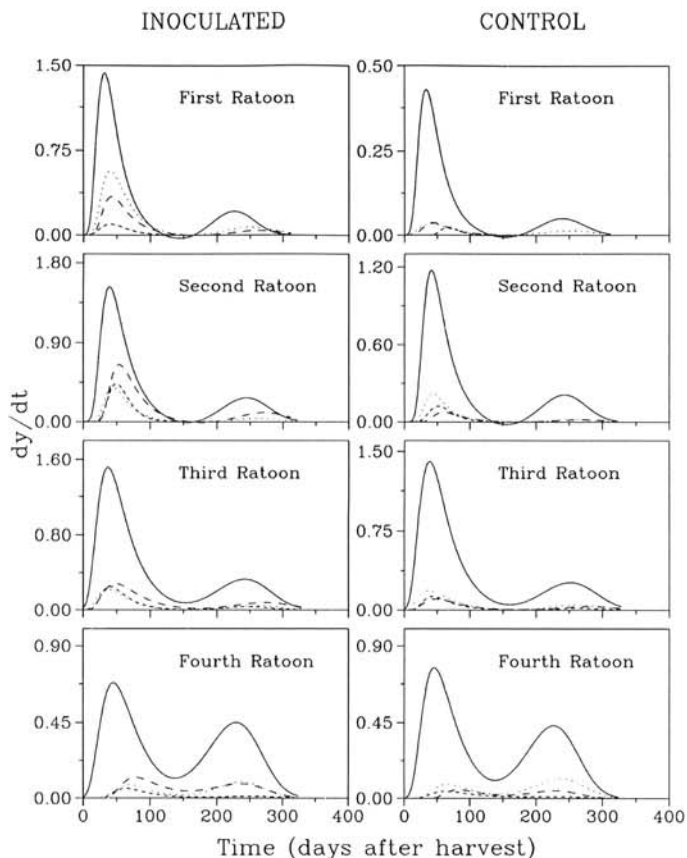
for SP71-799 in the first and second ratoon for both treatments and for SP71-1406 in the first ratoon for the control treatment. In these cases, the derivatives revealed only one wave, which was characterized by tp_1 and $y(tp_1)$; the other curve elements, tp_2, tp_3, and corresponding disease levels $y(tp_2)$ and $y(tp_3)$, did not exist. When two waves were expressed, the first wave was higher than the second wave in all cases.

None of the time-related curve elements, i.e., tp_1, tp_2, tp_3, and t_{max}, differentiated the cultivars, because the values of the curve elements were similar for all cultivars (mean values of 50, 160, 230, and 380 days for tp_1, tp_2, tp_3, and t_{max}, respectively) and were independent of the ratoon number and treatment. Disease levels at inflection points, i.e., $y(tp_1)$, $y(tp_2)$, and $y(tp_3)$, and upper asymptotes y_{max} clearly differentiated among cultivars (Fig. 4). The maximum disease level of cultivar NA56-79, the most susceptible cultivar, had significantly ($P < 0.05$) greater disease severity than the other three cultivars. Conversely, the cultivar SP71-799 had consistently the lowest values for y_{max}, differing statistically from the other cultivars. The level of resistance of cultivars SP71-1406 and SP71-6163, estimated by y_{max}, was intermediate. In all ratoons, y_{max} of SP71-6163 was slightly higher in the inoculated treatment but lower in the control treatment.

Rate functions, $r(t)$, had a minimum value about 150 days after harvest that corresponded to time tp_2 (Fig. 5). The curves for $r(t)$ obtained in the control treatment (not shown) were similar to those in the inoculated treatment. The mean infection rates ($\bar{r}$), determined by the integral of the rate function, were between 0.016 and 0.032 in the inoculated treatment and between 0.014 and 0.033 in the control treatment (Table 2). The functions for the rates of cultivar SP71-799 in the first and second ratoons were left out because the disease progress curve exhibited no second growth phase. Therefore, only higher values were calculated for progress curves.
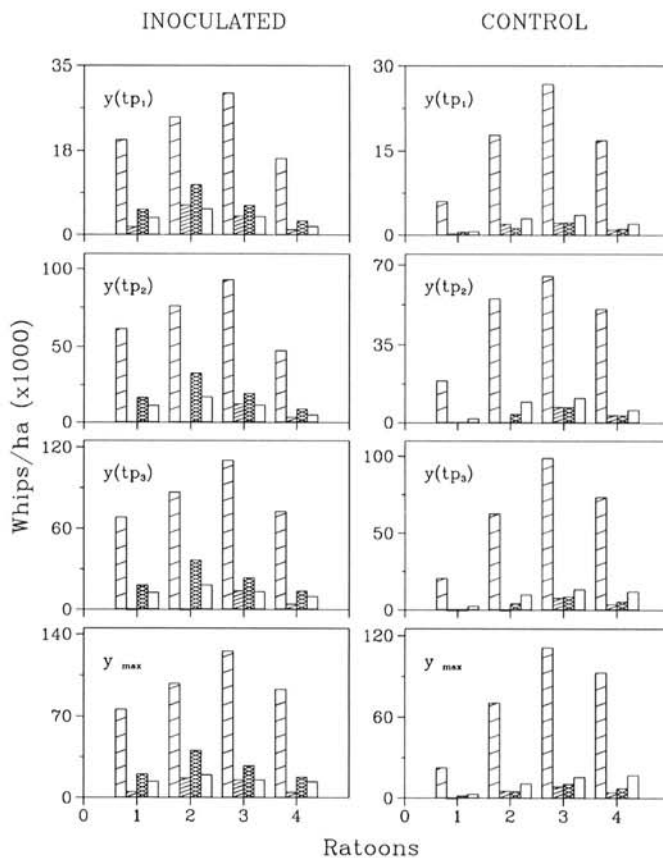

Fig. 3. Derivatives of the generalized Gompertz function fitted to smut data on the cultivars NA56-79 (—), SP71-799 (- -), SP71-1406 (– –), and SP71-6163 (···).

Fig. 4. Disease level elements ($y(tp_1)$, $y(tp_2)$, $y(tp_3)$, and y_{max}) of the smut progress curves for the cultivars NA56-79 (▨), SP71-799 (▨), SP71-1406 (▨), and SP71-6163 (□). Most standard deviations are between 0.5 and 2% of the estimated y, maximum value 9.8%. See text and Fig. 1 for details.
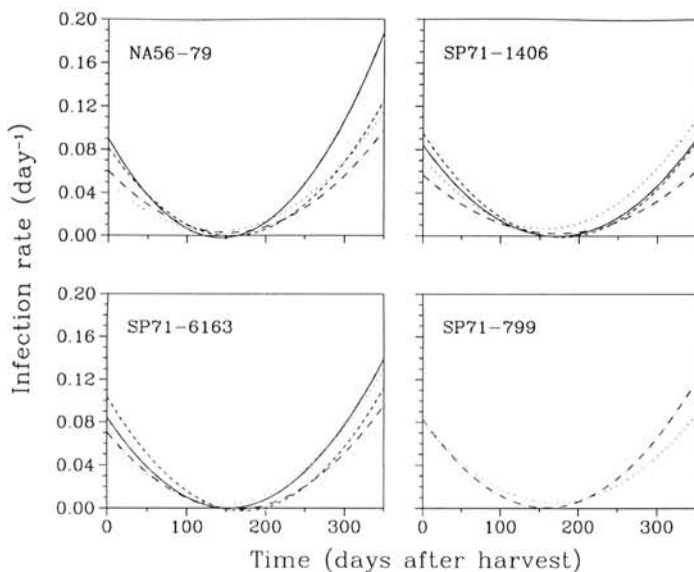
Fig. 5. Time function of the infection rate $r(t)$ obtained by the generalized Gompertz function for cultivars NA56-79, SP71-1406, SP71-6163, and SP71-799 in the first (—), second (- -), third (– –), and fourth (···) ratoons (treatment with dip inoculation).

TABLE 2. Mean infection rate ($\bar{r}$, days^{-1}), estimated by the generalized Gompertz function, for the disease progress curves of sugarcane smut on ratoons of four commercial sugarcane cultivars

Cultivar	Inoculated ratoons[a]				Control ratoons			
	First	Second	Third	Fourth	First	Second	Third	Fourth
NA56-79	0.032	0.024	0.023	0.026	0.026	0.026	0.020	0.027
SP71-799	0.040*[b]	0.068*	0.026	0.023	0.077*	0.059*	0.020	0.021
SP71-1406	0.020	0.019	0.016	0.027	0.025	0.019	0.014	0.033
SP71-6163	0.026	0.019	0.020	0.028	0.019	0.023	0.018	0.025

[a] Dip-inoculated with *Ustilago scitaminea*.
[b] Asterisks indicate values obtained when p_4 and p_5 were equal to 0.

DISCUSSION

The qualitative characteristics of progress curves for sugarcane smut, especially the presence of two growth waves, have been discussed by Amorim and Bergamin Filho (1). In this study, the quantitative analysis of these curves was described using a single mathematical function for the whole epidemic. Under this new approach, the annual disease progress curve was considered as a single process with a variable infection rate, which was a parabolic function of time (6). The generalized monomolecular and Gompertz functions fit the observed data with highly significant coefficients of determination (Table 1). Keep in mind that disease increase, at least in the initial phase, was not proportional to the amount of disease itself, but to the inoculum present in the soil.

Variation in shape of disease progress curves of monocyclic diseases was explored in depth by Jeger (8). It was evident that even in this type of disease, the monomolecular growth function only occurred when inoculum and rate of infection were constant. In other situations, more frequent in nature, sigmoid as well as asymptotic exponential curves may be observed (8). Therefore, the utilization of the generalized Gompertz function to fit sugarcane smut curves should not lead to inferences that relate to the type of development of the disease (12).

The progress curves of sugarcane smut were similar in shape but reflected epidemics of different intensities. Cultivar NA56-79,

the most susceptible cultivar, showed high smut severity in the inoculated and in the control treatments (Figs. 2 and 4). However, the difference in susceptibility between cultivar NA56-79 and the others was not reflected in the time-related elements of the progress curves, for instance, in tp_1, which can be defined as the latent period. The predicted latent periods were almost always close to 50 days, regardless of cultivar, ratoon, or treatment. In this pathosystem, latency was not a very useful parameter to differentiate cultivars. Usually, the latent period is a component of resistance capable of reducing the infection rate, especially in polycyclic diseases (11). In that case, the cumulative effect of a prolonged latency over several infection cycles of the pathogen may effectively reduce the disease growth rate. This was not the case with sugarcane smut, in which the fungus produced only two infection cycles in each annual cycle of the crop. The difference of a few days in the latent period of *U. scitaminea* did not really affect the course of the epidemic. Also, the mean infection rate $\bar{r}$ was not a very useful parameter to characterize smut resistance, although the infection rate is the usual parameter to differentiate cultivars in other pathosystems. In the four sugarcane cultivars compared, the values for the mean infection rates were similar (Table 2). Thus, neither time-related elements of the curves nor infection rates can be used to express differences in host resistance. The cultivars can only be differentiated by disease levels, especially by the maximum level of disease.

The application of the generalized Gompertz function in the sugarcane smut pathosystem, as proposed in this paper, allowed the description of the entire annual progress curve and permitted the determination of several parameters of epidemiological importance, such as average rate of infection, duration of the latent period, asymptote for the first growth phase, transition time from the first to the second growth phase, and final asymptote. The latter value is a well-suited and reliable parameter to quantify resistance of sugarcane cultivars to smut.

LITERATURE CITED

1. Amorim, L., and Bergamin Filho, A. 1991. Sugarcane smut development models: I. Annual curves of disease progress. Z. Pflanzenkrankh. Pflanzenschutz 98:605-612.
2. Antoine, R. 1961. Smut. Pages 326-353 in: Sugarcane Diseases of the World. J. P. Martin, E. V. Abbott, and C. G. Hughes, eds. Elsevier, Amsterdam.
3. Dixon, W. J. 1985. BMDP Statistical Software. University of California Press, Berkeley.
4. Draper, N. R., and Smith, H. 1981. Applied Regression Analysis. 2nd ed. John Wiley & Sons, New York.
5. Eisensmith, S. P. 1985. PlotIT Graphics and Statistics. ICS, Frankfurt.
6. Hau, B., Amorim, L., and Bergamin Filho, A. 1993. Mathematical functions to describe disease progress curves of double sigmoid pattern. Phytopathology 83:928-932.
7. Hau, B., and Kranz, J. 1990. Mathematics and statistics for analyses in epidemiology. Pages 12-52 in: Epidemics of Plant Diseases. Mathematical Analysis and Modeling. 2nd ed. J. Kranz, ed. Springer-Verlag, Berlin.
8. Jeger, M. 1987. The influence of root growth and inoculum density on the dynamics of root disease epidemics: Theoretical analysis. New Phytol. 107:459-478.
9. Kretschmann, H. J., and Wingert, F. 1971. Computeranwendungen bei Wachstumsproblemen in Biologie und Medizin. Springer-Verlag, Heidelberg.
10. Momol, M. T. 1986. Epidemiological studies of sugarcane smut. M.S. thesis. University of Florida, Gainesville.
11. Parlevliet, J. E. 1979. Components of resistance that reduce the rate of epidemic development. Annu. Rev. Phytopathol. 17:203-222.
12. Pfender, W. F. 1982. Monocyclic and polycyclic root diseases: Distinguishing between the nature of the disease cycle and the shape of the disease progress curve. (Letter to the editor.) Phytopathology 72:31-32.