

A Nonparametric Comparison of Monomolecular Growth Curves: Application to Western Painted Turtle Data

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Intractable distributional assumptions are generated in parametric analyses comparing groups of subjects described by nonlinear growth models. The distributional situation becomes more complex when pointwise mean curves best describe group growth (instead of curves based on mean parameters). An interval specific randomization test is developed to handle monomolecular growth models. The need for the technique was motivated by a study describing longitudinal growth data from the annuli on abdominal scutes of groups of western painted turtles. The analyses indicate that female turtles grow larger than male through the first seven years of life. The analytical procedures developed for these turtles are useful in the description and analyses of many other growth processes.

In studies of growth it is often desirable to describe the growth of individuals with mathematical models as a prelude to further describing and comparing growth patterns of groups of individuals.

Superb methodology and software exist for fitting and analyzing such curves for the case where the models are linear functions of the parameters, such as polynomial models, and where all stochastic components involved may be characterized as having multivariate normal distributions with particularly patterned covariance matrices. Groups are usually described and compared in terms of

group mean parameter vectors. Laird and Ware (1982) present an excellent overview of this parametric (multivariate normal) methodology for linear growth curve models.

The growth of living organisms is more often than not better described by nonlinear models, often sigmoid or S-shaped curves such as the Richards model (Von Bertalanffy, 1941; Richards, 1959; Draper and Smith, 1981). Parametric methodology appropriate for the analysis of nonlinear growth curves is discussed by White and Ratti (1977), White and Brisbin (1980), Sheiner and Grasela (1984), Beal (1984), and Brisbin *et al.* (1986a, 1986b). All

assume some variant of multivariate normality. Most describe and compare groups in terms of group curves defined by mean parameter vectors.

Ghosh *et al.* (1973), Zerbe (1979a, 1979b, 1979c), and Zerbe and Murphy (1986) describe nonparametric (distribution-free) methods appropriate for the analysis of nonlinear growth curves. While Ghosh's method describes and compares groups in terms of group curves defined by mean parameter vectors, Zerbe's method compares pointwise mean curves over an interval of time prespecified by the investigator.

In theory the nonparametric interval specific approach can be applied to growth curves of any form. In practice computational formulae have been available only for a class of growth curves which, unfortunately, excludes the Richards family of curves.

The objectives of this paper are to further develop the interval specific methodology as an alternative to standard parametric methodology for the analysis of nonlinear growth curves. Computational formulae are provided for the case where the growth curves are monomolecular, and the analysis is applied to the growth of the Western Painted Turtle.

THE USE OF NONPARAMETRIC GROWTH CURVE PROCEDURES FOR LINEAR GROWTH CURVE MODELS

The most obvious reason for using nonparametric (distribution-free) growth curve techniques is to avoid the usual assumptions of normality. But there are other reasons.

Most of the linear model growth curve literature concerns itself with distinguishing between residual variability of a subject's observations about that subject's regression line (within subject variability) and variability of subject parameter vectors (and hence curves) about mean parameter vectors (between subject variability). This leads to a mathematically neat analysis for the case where every subject is measured at the same ages (e.g., Rao, 1965; Grizzle and Allen, 1969), but further distributional difficulties when subjects are measured at different ages (Laird and Ware, 1982).

To fix ideas consider a linear growth curve model of the type discussed by Laird and Ware (1982). For subject i define the growth curve model,

$$Y_i = X_i B_i + e_i, \quad (1)$$

where Y_i is the vector of n_i growth measurements ordered by age, X_i is a design matrix, the elements of which are functions of time, B_i is a vector of parameters, and e_i is a vector of residual errors about the regression line. If the

growth curve for subject i was to be a straight line fit through three measurements, Y_{i1} , Y_{i2} , and Y_{i3} , recorded at ages, t_{i1} , t_{i2} , and t_{i3} , respectively, then (1) would be of the form

$$\begin{bmatrix} y \\ i1 \\ y \\ i2 \\ y \\ i3 \end{bmatrix} = \begin{bmatrix} 1 & t \\ & i1 \\ 1 & t \\ & i2 \\ 1 & t \\ & i3 \end{bmatrix} \begin{bmatrix} B \\ i0 \\ B \\ i1 \end{bmatrix} + \begin{bmatrix} e \\ i1 \\ e \\ i2 \\ e \\ i3 \end{bmatrix}$$

where B_{i0} is the intercept and B_{i1} is the slope of the subject's regression line. The error vector e_i is assumed to have a multivariate normal distribution with expectation vector 0 and covariance matrix R_i . This is the "within individual model" accounting for residual variation of an individual's observations about the individual's regression line.

To account for between subject variability, the subject's parameters are considered stochastic and permitted to vary from subject to subject according to a "between individual model." Let us assume a simple one,

$$B_i = \alpha + b_i, \quad (2)$$

where B_i is the subject parameter vector from (1), α is the mean parameter vector for the subject's group, and b_i is a random deviation vector assumed multivariately normally distributed with expectation vector 0 and covariance matrix D . It is further assumed that the within subject errors e_i and between subject errors b_i are independent.

Substituting (2) in (1) yields the "total mixed model,"

$$Y_i = X_i \alpha + X_i b_i + e_i, \quad (3)$$

or

$$Y_i = X_i \alpha + \epsilon_i, \quad (4)$$

where the total errors ϵ_i are multivariately normally distributed with mean vector 0 and covariance matrix $X_i D X_i' + R_i$.

Note that serial correlation may enter the total model either through the component R_i attributable to residual errors or through the component $X_i D X_i'$ attributable to differences between parameters of different subjects. Growth curve data is often measured so accurately and fit so well that the between subject component of the variation dominates the within subject component. As a consequence, serial correlation in the within subject model can often be ignored provided that the between component of variation is appropriately considered. Hence, it often facilitates analysis to assume that $R_i = I\sigma^2$. Laird and Ware (1982) refer to this as "conditional independence." Total errors may still be correlated and be heteroscedastic through the between component.

With the assumption of conditional independence, it is natural to fit the within individual model (1) to each subject separately, obtaining

$$\hat{B}_i = (X_i' X_i)^{-1} X_i' Y_i \quad (5)$$

for subject i 's parameter vector estimate. From the total model (4) the parameter vector estimates will have expectation vector α and covariance matrix $D+(X_i' X_i)^{-1} \sigma^2$. This is a problem. Unless every subject is measured at exactly the same n_i times, the design matrices X_i will vary from subject to subject and as a consequence every subject's parameter vector estimate will have a different covariance matrix. Then the common practice of comparing groups by treating each subject's parameter estimates as dependent variables in a multivariate analysis of variance (or even a univariate t test) becomes suspect since this second stage analysis requires that the covariance matrices of the dependent variables remain constant for all subjects in all groups being compared.

In order to compensate for this problem Laird and Ware (1982) introduce a complicated iterative procedure featuring the E-M algorithm in order to obtain maximum likelihood estimates of the group mean parameter vector α and likelihood ratio tests comparing such mean vectors for several groups. Their approach has the added virtue of fitting the total model (4) directly. It is unnecessary to fit individual models (1) for each subject. This feature is time saving and makes it possible to include subjects with single observations in the analysis.

As an alternative approach to the problem, Zerbe and Walker (1977) argue that the distributions of the estimated parameter vectors (5) have different covariance matrices because they are conditional on different times of measurement, and that it is reasonable to base inferences upon their unconditional distributions which will have constant covariance matrices, but which will in general not be multivariate normal. This suggests the use of distribution-free methods such as those proposed by Ghosh *et al.* (1973) and Zerbe and Walker (1977).

THE USE OF NONPARAMETRIC GROWTH CURVE PROCEDURES FOR NONLINEAR GROWTH CURVE MODELS

Nonlinear growth curve analysis has this same problem. The linear "within individual model" (1) is simply replaced by a nonlinear one, say

$$Y_i = F(B_i, t_i) + e_i, \quad (6)$$

where $F(B_i, t_i)$ is some nonlinear function of the parameter

vector B_i . F also depends on the times of measurement t_i . The "across individual model" may remain in the form (2). Substituting a linear across individual model (2) into a nonlinear within individual model (6) results in a total model,

$$Y_i = F(\alpha + b_i, t_i) + e_i, \quad (7)$$

which is nonlinear in the between subject errors b_i as well as nonlinear in the group parameters α . The resulting "total" distribution of Y_i is almost certainly nonnormal. Hence, parameter estimation and hypothesis testing based on the full likelihood of the data appears mathematically intractable, although Sheiner and Grasela (1984) and Beal (1984) have partially succeeded.

Let's consider an intuitive approach that sacrifices any information in the data Y_i not contained in the subject parameter vector estimates. Suppose that it is reasonable to assume conditional independence ($R_i=I\sigma^2$) and to fit the within individual model (6) to each subject separately, obtaining iteratively

$$\hat{B}_{i,j+1} = \hat{B}_{i,j} + (Z_i' Z_i)^{-1} Z_i' [Y_i - F(\hat{B}_{i,j}, t_i)] \quad (8)$$

for subject i 's parameter vector estimate, where $Z_i = \delta F / \delta B_i$ evaluated at $B_i = B_{i,j}$, $B_{i,0}$ is some initial guess of what B_i is, and $B_{i,1}, B_{i,2}, \dots, B_{i,j+1}$ are successive improvements of the estimate of B_i . Then it can be argued that the parameter vector estimates (8) will have approximately the expectation vector α and covariance matrix $D+(Z_i' Z_i)^{-1} \sigma^2$. Note that the Z_i vary from subject to subject, not only because the times of measurement t_i vary from subject to subject, but also because the parameters B_i vary from subject to subject. Then again, the common practice of comparing groups by treating each subject's parameter estimates as dependent variables in a multivariate analysis of variance (or a univariate t test) could be objected to because the second stage analysis requires not only multivariate normality, but that the covariance matrices of the dependent variables remain constant for all subjects in all groups being compared. One might argue in many growth curve situations that the within component of variability is small relative to the between component, and hence that the total covariance is nearly constant from subject to subject. In this situation one should also argue that it is not worthwhile to worry about serial correlation in the small within component.

The more scant nonlinear growth curve literature has not faced up to this problem as well as its linear growth curve counterpart. Too much emphasis has been placed on

accounting for possibly correlated, but small, residual errors and not enough on larger errors attributable to differences between subjects. Brisbin *et al.* (1986a, 1986b) treat parameter vector estimates obtained from nonlinear Richards models as dependent variables in second stage analyses of variance even though the homogeneity of covariance matrices that they require is not likely obtained. In an earlier paper White and Ratti (1977) did employ a nonlinear growth curve model somewhat analogous to the Laird and Ware (1982) model for linear growth curves, but unfortunately treated the differing subject parameters as fixed effects rather than more realistically as random effects.

Sheiner and Grasela (1984) and Beal (1984) are cognizant of the problem and have introduced a general nonlinear growth curve model that accounts for both between and within subject variability in a manner similar to that discussed by Laird and Ware (1982) for linear growth curves. Unfortunately, the complexity of the resulting distribution theory has forced them into some approximations that may seriously limit its usefulness. Nonetheless, their nonlinear mixed effects model (NONMEM) approach may be the soundest parametric approach currently available for the analysis of nonlinear growth curves.

Meanwhile the nonparametric approaches suggested by Ghosh *et al.* (1973), and Zerbe (1979a, 1979b, 1979c) are just as valid for comparing groups of nonlinear growth curves as they are linear growth curves. These authors differ principally in that the former compares group growth curves defined by parameters which are the means of the corresponding parameters for individual subjects, while the latter compares group growth curves which are the pointwise means of the individual curves within the groups.

THE USE OF POINTWISE MEAN CURVES

Whether it is worthwhile or not to fit individual growth curves with biologically interpretable parameters is an old debate. We will not take sides. We would like to point out, however, that even if the parameters of individual subject's growth curves are biologically interpretable, it does not follow that the means of these parameters are so easily interpretable. It is possible that a group growth curve defined by the means of the corresponding parameters for individual subjects may have no resemblance to any of the individual growth curves from the group it is supposed to represent. It is also possible that within some intervals of the age range of interest such a group growth curve may lie entirely above (or below) all of the individual curves. A pointwise mean curve will always lie in the midst of the individual curves and therefore will represent its group. They are not often used parametrically because the complexity of the resulting distribution theory would preclude

comparisons of pointwise mean curves based on multivariate normal theory. Fortunately the distribution-free approach makes such analysis feasible.

THE INTERVAL SPECIFIC RANDOMIZATION TEST

Incorporating concepts introduced by Fisher (1935), Welch (1937), Pitman (1937), and particularly Kempthorne (1952, 1955), Zerbe and Walker (1977) introduced a randomization test appropriate for the nonparametric comparison of two or more groups of growth curves over a specific interval of time. It was refined by Zerbe (1979a, 1979b) and extended to the problem of dynamically classifying the growth of individuals over a specified time interval by Zerbe (1979c) and Albert (1983).

The virtues of the interval specific randomization test are that it is nonparametric, age interval specific, compares pointwise mean curves, and is applicable to growth curves of any form, linear or nonlinear in the parameters. In fact, it is often used with growth curves generated by simple interpolation (e.g. Goldberg *et al.*, 1980; Albert *et al.*, 1982; Chapelle *et al.*, 1982; Hiatt *et al.*, 1983).

To briefly describe the interval specific randomization test, consider the one way analysis of variance model,

$$Y_{ij}(t) = \mu(t) + \tau_i(t) + \epsilon_{ij}(t), \quad (9)$$

where $Y_{ij}(t)$ is a growth curve estimated for subject j in group i , $\mu(t)$ is a grand mean growth curve, $\tau_i(t)$ is an effect curve for group i , and $\epsilon_{ij}(t)$ is an error curve associated with subject j in group i . No distribution assumptions are required of the $\epsilon_{ij}(t)$ and no particular form is required of the estimated growth curves $Y_{ij}(t)$. Zerbe and Walker (1977) proposed testing the interval specific null hypothesis that

$$\tau_1(t) = \tau_2(t) = \dots = \tau_g(t) = 0 \\ \text{for all } t \in (a,b),$$

i.e., that the treatment effect curves are all zero throughout some age interval of interest (a,b) , with the statistic,

$$F = [B/(n-1)] / [W/(n-g)],$$

where the between (B), within (W), and total (T) sums of squares integrated over the time interval (a,b) are defined in Table 1.

Based on the statistical model (9) for random growth curves Zerbe (1979a) showed that the null permutation distribution of F could be well approximated by a standard F distribution with certain synthesized degrees of freedom, thus avoiding the necessity of costly recomputation of F for every reassignment of the subjects. It has been demon-

TABLE I

Completely Randomized Analysis of Variance of Growth Curves Over the Time Interval (a,b)

Source	Sum of Squares	D.F.	Mean Square	Expected Mean Square
Between	$B = \sum_i n_i \int_a^b \{\bar{y}_{i.}(t) - \bar{y}_{..}(t)\}^2 dt$	$g-1$	$B/(g-1)$	$\int_a^b \sigma^2(t) dt + (g-1)^{-1} \sum_i n_i \int_a^b \{\tau_i(t) - \bar{\tau}_{.}(t)\}^2 dt$
Within	$W = \sum_{ij} \int_a^b \{y_{ij}(t) - \bar{y}_{i.}(t)\}^2 dt$	$n-g$	$W/(n-g)$	$\int_a^b \sigma^2(t) dt$
Total	$T = \sum_{ij} \int_a^b \{y_{ij}(t) - \bar{y}_{..}(t)\}^2 dt$	$n-1$		

Source: Zerbe, Gary O. (1979). Randomization of the completely randomized design extended to growth and response curves. *Journal of the American Statistical Association* 74, 215-221.

strated that the interval specific test is more powerful than any of its nonparametric competitors (Krause-Steinrauf, 1986) for vertical shift alternatives to the null hypothesis. Also Foutz *et al.* (1985) and Zerbe and Murphy (1986) have recently tackled multiple comparison problems associated with the procedure.

Zerbe (1979a) provided convenient computational algorithms and Nelson and Zerbe (1988) provided SAS compatible software for executing the analysis for a class of growth curves that included polynomials, moving averages, and linearly interpolated curves as special cases. Unfortunately, nonlinear growth curves, such as the Richards family of curves were not members of this class.

DEVELOPMENT FOR COMPARISON OF MONOMOLECULAR GROWTH CURVES

The computational procedures provided by Zerbe (1979a) to cover a broad class of functional forms of growth curves could not be directly applied to monomolecular growth curves. We develop an analysis for monomolecular growth curves beginning with computational formulae for Zerbe's between (B), within (W), and total (T) sums of squares below:

$$B = \sum_i n_i \int_a^b \bar{y}_{i.}^2(t) dt - n \int_a^b \bar{y}_{..}^2(t) dt \quad (10)$$

$$W = \sum_{ij} \int_a^b y_{ij}^2(t) dt - \sum_i n_i \int_a^b \bar{y}_{i.}^2(t) dt \quad (11)$$

$$T = \sum_i \sum_j \int_a^b y_{ij}^2(t) dt - n \int_a^b \bar{y}_{..}^2(t) dt. \quad (12)$$

Computation of the above sums of squares requires the following three quantities:

$$\int_a^b y_{ij}^2(t) dt = q(i, j, i, j) \quad (13)$$

$$\int_a^b \bar{y}_{i.}^2(t) dt = (1/n_i^2) \sum_j \sum_{j'} q(i, j, i, j') \quad (14)$$

$$\int_a^b \bar{y}_{..}^2(t) dt = 1/n^2 \sum_i \sum_j \sum_{i'} \sum_{j'} q(i, j, i', j') \quad (15)$$

where

$$q(i, j, i', j') = \int_a^b y_{ij}(t) y_{i'j'}(t) dt. \quad (16)$$

For monomolecular growth curves of the form

$$Y_{ij}(t) = A_{ij} + B_{ij} \exp(-G_{ij}t),$$

$$q(i, j, i', j') = \int_a^b [A_{ij} - B_{ij} \exp(-G_{ij}t)]$$

$$[A_{i'j'} - B_{i'j'} \exp(-G_{i'j'}t)] dt$$

$$= \left[\begin{array}{l} A_{1,j}A_{1,j} + \frac{A_{1,j} \cdot B_{1,j} \exp(-G_{1,j}t)}{G_{1,j}} \\ + \frac{A_{1,j}B_{1,j} \cdot \exp(-G_{1,j} \cdot t)}{G_{1,j}} \\ - \frac{B_{1,j}B_{1,j} \cdot \exp(-G_{1,j}t - G_{1,j} \cdot t)}{G_{1,j} + G_{1,j}} \end{array} \right] \left| \begin{array}{l} b \\ a \end{array} \right.$$

The degrees of freedom, v_1 and v_2 , for Zerbe's approximate F test may be calculated as

$$v_1 = 2E^2(1-E)/V - 2E$$

and

$$v_2 = (1-E)v_1/E$$

where

$$E = (g-1)/(n-1),$$

$$V = [(r-s)p^4 + qk^2]/\sigma^4 + s,$$

and in turn,

$$e = \Sigma_1(1/n_1),$$

$$m = (n-1)^2(n-2)(n-3),$$

$$q = [n(n+1)e - (g^2+2g-2)n+g(g-2)]/m,$$

$$r = [2(g-1)n^3 - 3n(n-1)^2e + (g^2-4g+6)n^2 - 6n-3g(g-2)]/(nm),$$

$$s = [-n(n-1)^2 + g^2n^2 - 2(g^2-g+1)n - g(g-2)]/(nm),$$

$$\sigma^2 = T/(n-1),$$

and p^4 and k^2 may be determined from:

$$(n-1)p^4 = \Sigma_1 \Sigma_j \Sigma_{i'} \Sigma_{j'} [q(i,j,i',j') - (1/n)\Sigma_1 \Sigma_j q(i,j,i'',j'') - (1/n)\Sigma_1 \Sigma_j q(i',j',i''',j''')] + (1/n^2)\Sigma_1 \Sigma_j \Sigma_{i''} \Sigma_{j''} [q(i'',j'',i''',j''')]^2$$

and

$$(n-1)k^2 = \Sigma_1 \Sigma_j [q(i,j,i,j) - (2/n)\Sigma_1 \Sigma_j q(i,j,i'',j'')]$$

$$+ (1/n^2)\Sigma_1 \Sigma_j \Sigma_{i''} \Sigma_{j''} [q(i'',j'',i''',j''')]^2.$$

A computer algorithm was developed to do all of the above calculations and is available to the public (Rickard *et al.*, 1988).

APPLICATION TO GROWTH OF WESTERN PAINTED TURTLES

Western painted turtles were collected 30 August to 10 September, 1976, and 26 June to 2 July, 1978, at Medical Lake Washington. The study of the growth of these turtles is considered important for several reasons. First, the painted turtle is the most geographically widespread turtle in the United States, but no growth information has been reported in the northwestern most part of its range. Second, Medical Lake offers a rich environment for turtles; therefore, turtles from this location may achieve maximal growth for the species. Third, just prior to the second collection of turtles, the lake was "rehabilitated" (by reducing its alkalinity) for the introduction of trout. Describing the growth of these turtles provides a baseline for studying the effects of altering the environment on the turtles, which are important animals at the top of the food chain in the lake. Lastly, techniques for describing turtle growth would be useful where other, perhaps endangered species, are involved in habitat alterations.

We defined four groups of turtles for which we obtained growth data: males 1976, females 1976, males 1978, females 1978. A record of a turtle's growth history is contained in the annuli on the scutes: plates covering the turtle's shell (Ernst and Barbour, 1972; Gibbons, 1976, 1987). Each growing season, the germinal layer of epidermis produces an annulus on each scute analogous to a tree ring. The distances between the annuli on an individual abdominal scute provide a number of years of longitudinal growth data which we describe for each group and compare among groups. Growth in painted turtles has appeared in a variety of studies including Ernst (1971), MacCulloch and Secoy (1983), Sexton (1959) and Wilbur (1975).

The accuracy of estimating age and constructing growth curves for annuli measurements is based on several assumptions. First, a perceptible growth increment is formed each year. It is also assumed that accompanying this growth increment is the formation of only one annulus. Although it has been reported that during the first growing season young turtles often form one or two accessory annuli, these false annuli can be identified because they are not as distinct as true annuli. Last, we assume that the earliest rings disappear first, as reported by Gibbons (1987).

Initial attempts at describing the growth data were made using polynomial curves to describe the growth of individuals and mean curves (obtained by averaging parameters) to describe the four groups. These polynomial curves were

inadequate for describing observed growth because unrealistic decreases in size were predicted as turtles reached maturity.

Frequently, the Richards family of exponential models works well to describe growth (e.g. Goldstein, 1979; Draper and Smith, 1981; Brisbin *et al.*, 1986a, 1986b), and the behavior of the monomolecular curve, $Y = A - Be^{-Gt}$, was very similar to the plots of the individual turtle data. The model,

$$Y_{1jk} = A_{1j} - B_{1j}e^{-G_{1j}t_{1jk}} + e_{1jk},$$

$$k = 1, 2, \dots, n_{1j}, \quad (17)$$

was fit to the observations of each individual separately using the SAS procedure NLIN (SAS User's Guide: Statistics, 1982). The independent variable t_{ijk} represents the age in years and the dependent variable Y_{ijk} is the length of the

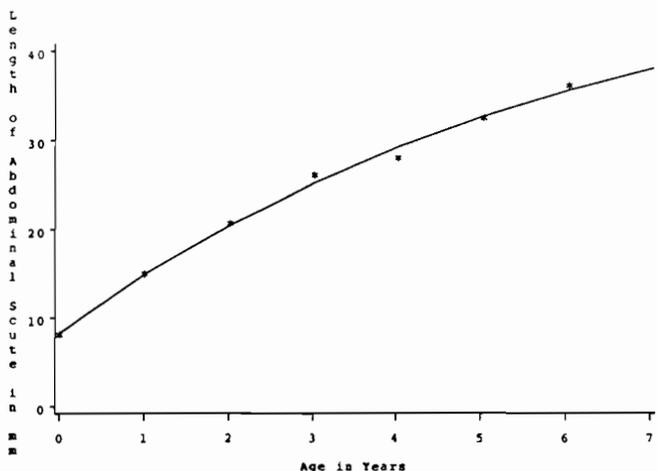
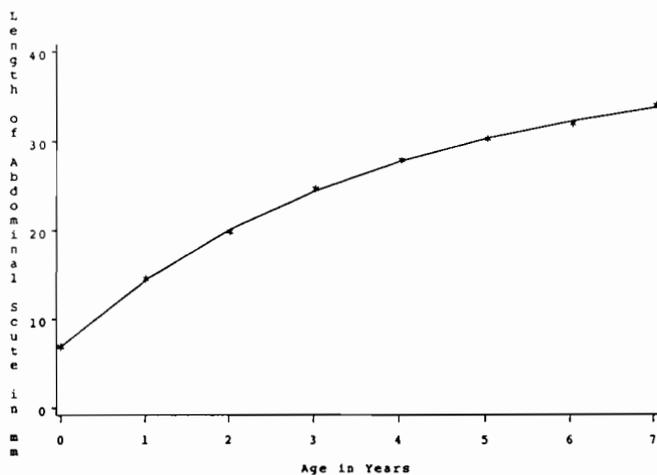


FIGURE 1. Original data overlaid by individual exponential curves for two randomly chosen turtles.

abdominal scute for the k th observation on turtle j in group i . The e_{ijk} are assumed uncorrelated random error terms with zero expectations and variances σ_{ij}^2 . This monomolecular model visually fit the data exceptionally well (Figure 1); so well, that it would be unreasonable to complicate the analysis by investigating possible serial correlation among the very small residuals. Any serial correlation associated with the larger between subjects component of variation is accounted for in the analysis.

Since the derivative, BGe^{-Gt} , of the monomolecular growth curve was of the same monomolecular form, the same methodology could be used to examine the velocity of turtle growth.

Attempts to fit the more general four parameter Richards model,

$$Y = [A^{1-m} - (A^{1-m} - C^{1-m})e^{-(1-m)Gt}]^{1/(1-m)},$$

failed. The model was over parameterized, resulting in questionable convergence and unstable parameter estimates with enormous standard errors. Since the instability was associated with the parameter m , and since we were obtaining such good fits with the monomolecular model ($m=0$), we abandoned the more general model.

Although the monomolecular model (17) proved adequate for individual data, the resulting group curves generated by averaging over the parameters tended to lie above the individual curves in the group and therefore were considered inadequate for describing the observed growth (Figure 2). Alternatively, taking pointwise means of the raw data would have lost a great deal of information because: (1) the sample sizes for each group were small, and

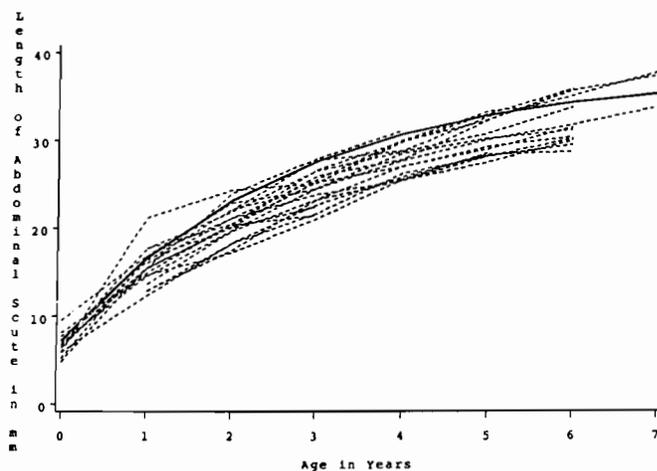


FIGURE 2. Example of inadequate description of group growth (females, 1978) generated by averaging the parameters from individual exponential curves.

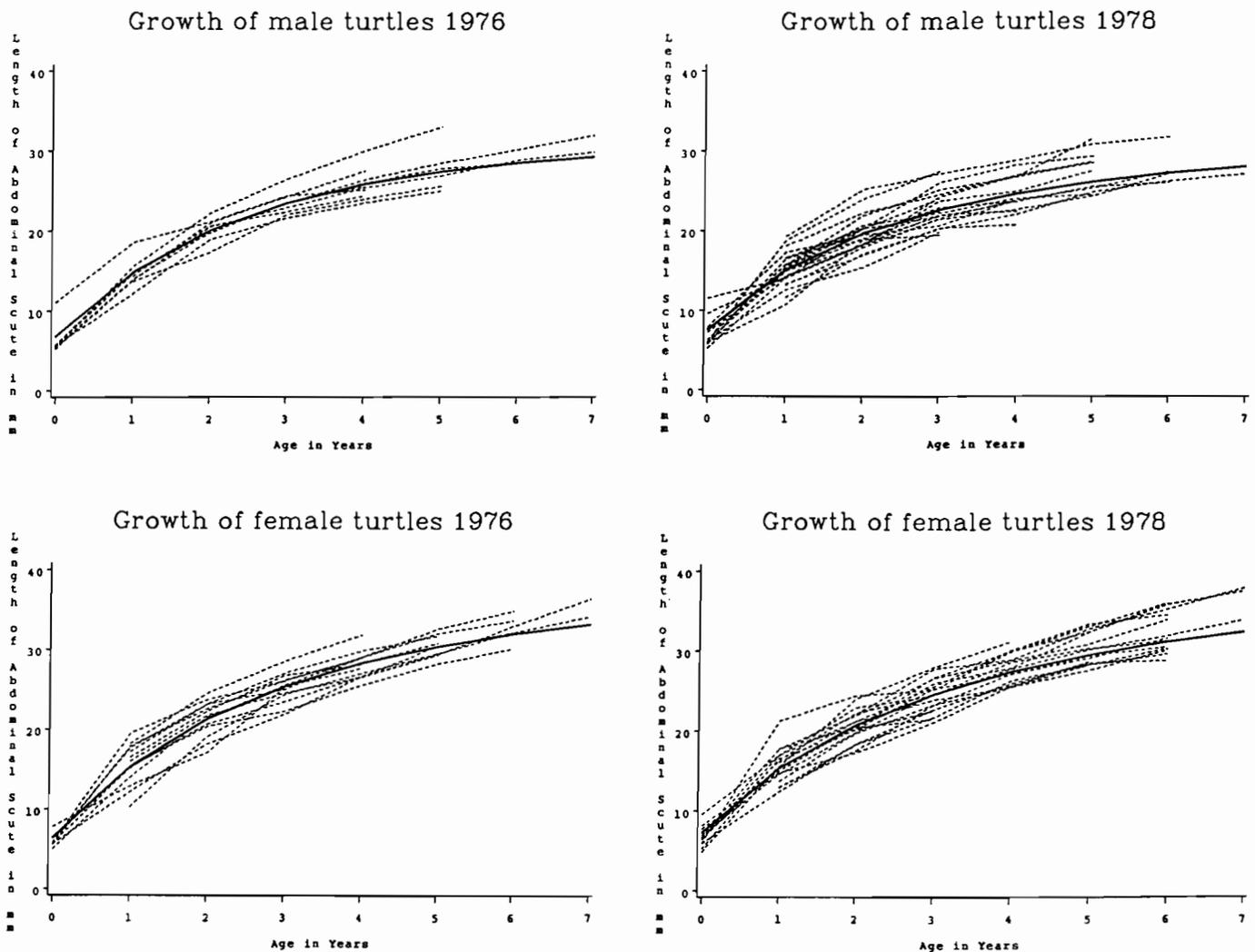


FIGURE 3. Original data overlaid by pointwise mean exponential curves.

(2) means constructed at many of the time points would have been based on only a few individuals.

Finally, we used pointwise averages of the exponential curves fitted to the individuals to describe growth of a group. The pointwise mean curves were consistent with the general characteristics of the raw data for each group (Figure 3). Because the data within each group were observed over different time segments within the range of interest (0 to 7 years), some individual curves had to be extrapolated before taking the pointwise means. Validation of the extrapolation procedure was accomplished by first determining the range of data time points for the turtle with the least data in each group. Using only data within that minimum range for the group, each turtle's data was refit to equation (1). All of these refitted curves were extrapolated

over the entire range of data and were graphically compared to the actual values in the extended range. These results indicated that, even under the worse case scenario above (minimal data points for fitting and extrapolating curves), the procedure of extrapolating curves of turtles with fewer data points to a larger range would pose no problems for use in calculating pointwise mean curves.

There were three comparisons of interest among the groups. The sexes each have different survivals, hence, growth between sexes should be compared. Growth curves for turtles captured in 1976 were compared to those for turtles captured in 1978 to see if there was any difference between cohorts. Any change in growth could have been due to the rehabilitation of the lake during the prior year. No turtle was in both annual cohorts.

RESULTS

No differences in growth curves could be detected between 1976 and 1978 within either sex (Table 2). Finding no differences among the cohorts indicated that the effects of the chemical treatments in 1977 (which would have affected only a couple of months of the 1978 growing season) did not have a detectable influence on the growth of the turtles in our sample. It also justified combining the two years of data for purposes of comparing sexes.

TABLE 2

Analytical Results from the Growth and Velocity Curve Comparisons

Hypotheses Tested	P-value for Growth Comparisons	P-value for Velocity Comparisons
females 78 = females 76	.410	.493
males 78 = males 76	.340	.282
males = females (overall)	.001	.010

Pointwise mean curves were constructed for the data from the two male groups combined and the two female groups combined. The female curve was significantly higher than the male curve ($p < 0.001$) over the age interval 1 to 7 years.

The same group comparisons were conducted for velocity curves as for the growth curves. These results are essentially the same as for the growth curve comparisons (Table 2). We combined the two year groups within each sex and computed an overall comparison of the sexes for the velocity curves. Females again were significantly higher ($p < 0.010$).

The consistent differences in growth between the sexes probably reflect different reproductive strategies. When turtles such as the painted turtle become sexually mature, their annual growth is drastically reduced (Gibbons, 1967; Wilbur, 1975; MacCulloch and Secoy, 1983). Wilbur (1975) argues that the size at which males become mature (which is smaller than that of females) is balanced between the advantages of being able to reproduce sooner versus the advantages of delaying reproduction in order to increase body size. Females mature at older ages than males, and the increased body size allows larger clutch sizes. Rapid juvenile growth decreases vulnerability to predators.

The present study utilized the increase in length of an abdominal scute to construct growth curves. These mea-

surements are generally transformed to plastron lengths in the turtle growth literature. This transformation is based on works by Sergeev (1937) stating that the relationship of the abdominal scute to the plastron remains approximately constant in painted turtles. Growth curves in this study were constructed using the original measurements to avoid potential bias and variation resulting from this transformation.

DISCUSSION

Our methodology is more appropriate for comparing monomolecular growth than the procedures suggested in the current literature which compare nonrepresentative curves generated from mean parameter vectors. Moreover, our methodology is nonparametric, not requiring multivariate normality and homogeneity of variance assumptions which can not possibly hold for parameter estimates generated from nonlinear models fit to subjects with varying times of observation. Also the procedure is interval specific, allowing the investigator to focus on the age interval of major interest. Judicious comparison over several prespecified age intervals may sharpen the growth curve analysis in much the same manner as a priori contrasts resolve an ordinary analysis of variance.

The methods developed here can help determine the effects that rehabilitation of Medical Lake has on the growth of its turtle population when further data are collected. A more general use of these growth curve methods would be to compare growth data in this study to growth data for other turtle populations. Examination of the growth and velocity curves for specific populations and comparisons between populations should add to the understanding of the growth process in turtles and other animals. We are currently considering an application of these techniques to growth data from a species for which some populations are considered threatened and are the focus of land use controversies.

Another current application is to describe and compare animal damage problems in crops. The growth of damage (loss) in a crop over a growing season may be modeled by a monomolecular growth curve. The efficacy of treatments could be compared using damage growth and velocity curves and may indicate the optimal time to treat a crop to minimize total damage by harvest time.

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