

## Multiple Regression Analysis of Aquaculture Experiments Based on the "Extended Gulland-and-Holt Plot": Model Derivation, Data Requirements and Recommended Procedures\*

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### Abstract

A method for the multivariate analysis of fish growth in aquaculture is presented. It is derived from a linearized version of the von Bertalanffy growth function (VBGF), which, in its original form, is a bivariate regression termed the Gulland-and-Holt plot. Here, a version in form of a multiple regression equation is presented. The "extended Gulland-and-Holt plot" permits to identify and quantify the key variables controlling fish growth and permits the inclusion of these environmental and treatment variables to explain variance in growth of fish. Von Bertalanffy growth parameters  $K$  and  $L_{\infty}$  are obtained, which contain the combined environmental effects on fish growth and reflect the range of culture conditions. By computing the index of growth performance ( $\phi$ ), the obtained regression models can be used for growth prediction and decisionmaking in fish farm management and production under a wide range of environmental and management conditions. Recommendations for the design of experiments, preparation of data for analysis and actual analysis procedures are given.

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### Introduction

For aquaculturists, the processes of main interest are fish growth and production (Bardach et al. 1972; Steffens 1981; Hepher and Pruginin 1981). When conducting experiments, the normal procedure is to compare the length of time needed to reach "market" sizes (i.e., length or weight), growth rates (e.g.,  $g \cdot day^{-1}$ ) or "production" (actually: standing stock in, e.g.,  $kg \cdot ha^{-1}$ ) at the end of a growth period. This requires that all variables are kept constant except for the treatments under investigation. Influences on the growth process itself during experiments are neglected. Several problems arise here:

1. Only one treatment at a time can be directly compared. Possible associated effects must be neglected. In ordinary growth experiments the final weights are compared using ANOVA or t-tests. This requires complete control of all variables and equal fish sizes at the beginning. The information contained in the growth curve itself is lost since only the single last value is used for interpretation.

For example, in single-factor experiments with a simple ANOVA table for analysis, only one variable can be compared (e.g., final fish sizes or average growth rates) and all that can be inferred is whether a positive or negative effect was observed. The disadvantage is that this method does not permit to quantify how much better one type of treatment was compared to the other. Further, associated effects among the variables are neglected, so that they remain undiscovered.

2. Uncontrollable and varying effects such as those caused by meteorological factors (seasonality of temperature and rain, environment, etc.) and different experiment locations are difficult to account for in the analysis.
3. The growth of fish is not linear. Thus, it is important to have (a) identical fish sizes at the beginning of the experiment, and (b) equal duration of experiments.

For the aquaculture scientist it is important to know the growth performance of certain species for the planning of aquaculture projects, the

development of industries at new locations, and the correct management of farms during different seasons or conditions (Reay 1979).

When conducting large experiments with many treatments (i.e., multifactor experiments), the problems mentioned above are worsened (Gomez and Gomez 1984). The amount of data increases exponentially with every further variable measured (Hopkins et al. 1988). Data handling and management become very complicated. With the ordinary methods of data analysis, the interpretation of results and the testing of hypotheses is very difficult. Today, the availability of computers with data management and statistical analysis software can help to solve many of these problems (Vakily 1989) even with sophisticated methods of data analysis. An approach along this line is a method developed by Pauly and Hopkins (1983, see Appendix I, this vol.) which will be explained in detail here. This may be regarded as representing only the beginning of this type of analysis. In the future, more methods of statistical analysis

and modeling will be developed since aquaculture is undermathematized compared to agriculture or fisheries.

### The Process of Growth

Curves describing growth in length or in weight both approach an asymptotic value towards the end of an animals' life span. Length growth can usually be modelled using an asymptotic curve which tapers off with increasing age. Weight growth is roughly sigmoid, i.e., the weight increment increases gradually up to an inflection point from where it then gradually decreases again. The growth rates change constantly, which imposes problems when using these parameters for interpretation of experiments (Fig. 1). For different fish sizes the absolute growth increments will be different (Fig. 1).

A method for the mathematical description of fish growth is the von Bertalanffy growth function

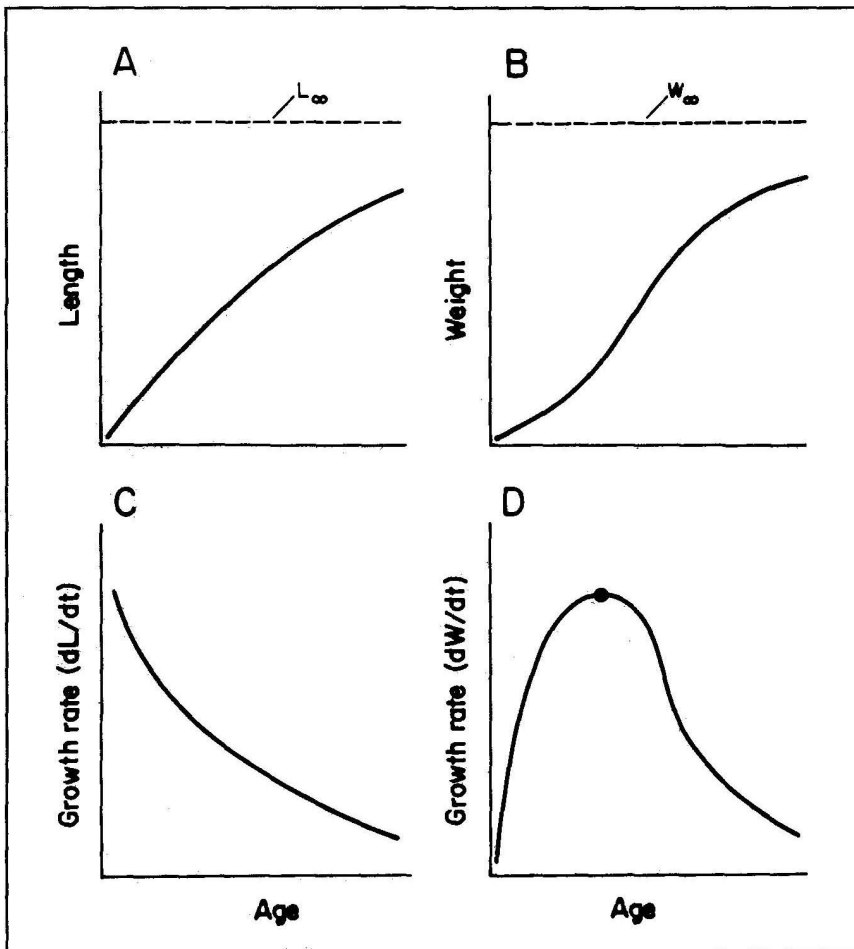


Fig. 1. Schematic example of growth curves. A) for length, and B) for weight, which has an inflection point. Both curves approach asymptotic size. C) Schematic representation of the change of absolute growth rate with time in length and D) in weight. Note the position of the inflection point in weight growth rate. The growth rate in weight of fish at different ages (sizes) cannot be compared.

(VBGF) (Pütter 1920; von Bertalanffy 1934, 1938), which is widely used in fisheries research. This function has several advantages as will be shown later. The VBGF, as modified by von Bertalanffy by Beverton and Holt (1957) is:

$$L_t = L_\infty \cdot \left(1 - e^{-K(t-t_0)}\right) \quad \dots 1$$

for length, and

$$W_t = W_\infty \cdot \left(1 - e^{-K(t-t_0)}\right)^m \quad \dots 2$$

for weight, where

$K$  = growth constant

$L_\infty$  = asymptotic length, i.e., the (mean) length the fish would reach if they were to grow indefinitely

$W_\infty$  = asymptotic weight, i.e., the weight corresponding to  $L_\infty$

$t$  = age of the fish in days/months/years

$t_0$  = theoretical (generally negative) "age" of the fish at zero size

$m$  = exponent of length-weight relationship

For the case of isometric growth, i.e., when  $m = 3$ , the VBGF takes the form:

$$W_t = W_\infty \cdot \left(1 - e^{-K(t-t_0)}\right)^3 \quad \dots 3$$

With these parameters the growth of a single fish or the mean of a whole fish population can be described. For reasons to be shown later we will concentrate on the length-based version of the VBGF.

### Estimation of VBGF Parameters

#### GULLAND-AND-HOLT PLOT

From aquaculture experiments the VBGF growth parameters  $L_\infty$  and  $K$  can be estimated, among several other methods, by a method termed the 'Gulland-and-Holt plot' (Gulland and Holt 1959), a method already described in von Bertalanffy's original derivation of his growth equation (von Bertalanffy 1934). This provides an approximative method to estimate the VBGF parameters by a simple linear regression technique. Here growth rates in length are regressed upon their corresponding average lengths during the intervals. The intervals need not be of equal duration. Also, different fish sizes and growth rates can be used.

This method is used in fisheries research for the analysis of tagging data and of length-frequency data (Gulland 1967, 1983; Pauly and Ingles 1981; Pauly 1984; Sparre et al. 1989). In aquaculture experiments we are in a situation similar to a tag-recapture experiment. Initially the ponds are stocked. During the experiment the fish are sampled periodically and finally the population is harvested. The Gulland-and-Holt method is based entirely on length measurements. Unfortunately this is in contrast to the procedure in most aquaculture experiments where only weights are measured.

The length growth curve is characterized by a gradual increase in length over time and a gradual decrease in growth rate (indicated by the slope of the tangent) over time. As length ( $L$ ) approaches its maximum (i.e.,  $L_\infty$ ) the growth rate becomes zero. This relationship between length and growth rate is linear and thus can be used to estimate the two parameters  $L_\infty$  and  $K$ . Growth rate ( $\Delta L/\Delta t$ ) of an experiment interval is plotted over the mean length in that interval. It should be noted that this is a linear description of a nonlinear growth process (Fig. 2). The differential form is:

$$dL/dt = K(L_\infty - \bar{L}) \quad \dots 4$$

or, in terms of growth increments per interval (length  $L_1$  and  $L_2$ ):

$$(L_2 - L_1)/(t_2 - t_1) = a + b(L_1 + L_2)/2 \quad \dots 5$$

It should be noted that (4) is a differential equation and (5) is a difference equation, yet both

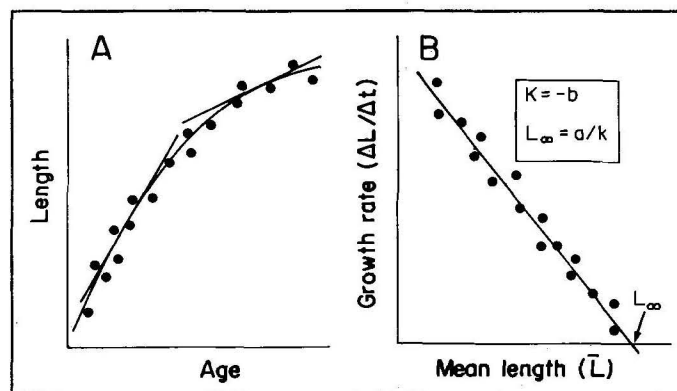


Fig. 2. A) Example of a length growth curve through data points. Tangents indicate the decrease in slope with age (i.e., size). B) Representation of a Gulland-and-Holt plot, describing the decrease in growth rate with increasing size. The VBGF parameters  $K$  and  $L_\infty$  are derived from the regression coefficients.

give similar results for small intervals. The growth parameters are obtained according to:

$$K = -b \quad \dots 6)$$

and

$$L_{\infty} = a/K \quad \dots 7)$$

Gulland and Holt (1959) pointed out that the discrepancy between difference and differential equations comes into effect at larger time intervals (i.e., several months to years). Therefore they suggested a correction factor with which the y-values are to be multiplied before computing the regression. For short intervals of 14 days this factor is very close to unity and can consequently be neglected (e.g., for  $K = 0.01482 \text{ day}^{-1}$  and  $\Delta t = 14$  days, the correction factor is 1.0033).

#### CONFIDENCE LIMITS FOR K

Since  $K = -b$ , the confidence limits (CL) for K, derived with the Gulland-and-Holt plot, are the same as those for 'b', only with the sign changed (Sparre et al. 1989):

$$CL_{(K)} : [K - s_{(K)} \cdot t(n-2), K + s_{(K)} \cdot t(n-2)] \dots 8)$$

where

- $CL_{(K)}$  = confidence limits of K
- $s_{(K)}$  = standard deviation of K (here with n-2 df)
- $t(n-2)$  = fractiles of Student's t-distribution, here based on n-2 degrees of freedom

#### CONFIDENCE LIMITS FOR $L_{\infty}$

According to Sparre et al. (1989) the confidence limits for  $L_{\infty}$  derived with the Gulland-and-Holt plot cannot be straightforwardly derived. Rather they are "conditioned" on a previously determined value for K together with the confidence limits for 'a' in form of a ratio. The confidence limits for 'a' are obtained using:

$$CL_{(a)} : [a - s_{(a)} \cdot t(n-2), a + s_{(a)} \cdot t(n-2)] \dots 9)$$

where  $s_{(a)}$  = standard deviation of 'a' (here with n-2 df).

Since  $L_{\infty} = -a/b$ , the procedure suggested by Sparre et al. (1989) is to divide the lower and upper confidence intervals of 'a' by the value of K:

$$CL_{(a)} : [(a - s_{(a)} \cdot t(n-2))/K, (a + s_{(a)} \cdot t(n-2))/K] \dots 10)$$

where  $CL_{(L_{\infty})}$  = confidence limits for  $L_{\infty}$ .

Sparre et al. (1989) point out that, strictly, the confidence limits of a ratio are not defined. An alternative method is to regard  $L_{\infty}$  as a predicted value of y (here zero), estimated from a given x-value (here the x-intercept). According to Snedecor and Cochran (1982), it is possible to estimate the confidence interval for  $L_{\infty}$  (in form of the x-axis intercept), as also suggested for the "extended Bayley plot" in Prein and Pauly (this vol.).

#### CONSIDERATIONS FOR APPLICATION

For fish size data based on weight, the weight values must be converted to lengths, using a length-weight relationship. Therefore, a length-weight relationship should always be determined for the fish stock used in the experiments, or a set of length-weight parameters should be used, estimated from a stock of fish kept under similar conditions.

When dealing with several experiments, a separate growth curve could be derived for each experiment, producing a range of combinations of VBGF parameters (Fig. 3). The VBGF parameters could then be related to the different treatments.

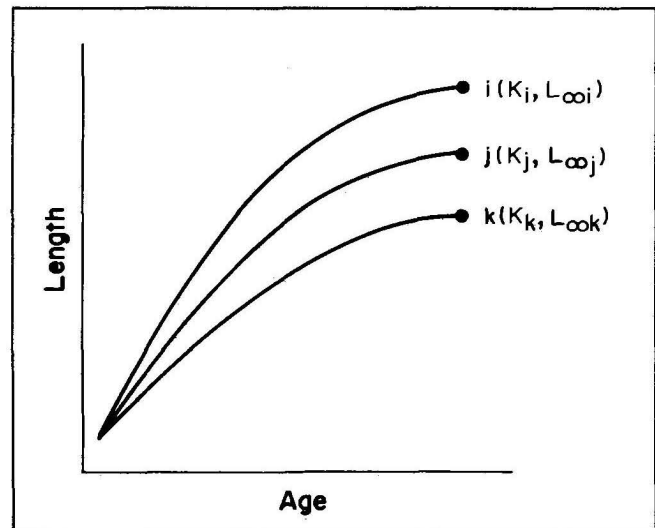


Fig. 3. Schematic example of fish growth (in length) under three different treatments (I, II and III). Usually, only sizes at the end of experiments, or overall yields, are compared (given equal sizes at stocking). In this way, any variations in growth occurring during the culture period are neglected. One possibility is to estimate a set of VBGF parameters for each pond or treatment.

The parameter combinations thus obtained could then be used for the prediction of growth under the assumption that all environmental and treatment factors are constant during the prediction range.

From the obtained pairs of VBGF growth parameters  $K$  and  $L_{\infty}$  the index  $\phi'$  (Pauly 1979; Pauly and Munro 1984) can be computed to compare the growth performance of different fish stocks. It has been shown that the index of growth performance  $\phi'$  can be used to compare the growth of tilapias under a wide range of culture conditions, within and between species and between sexes (Pauly et al. 1988). The growth performance index is computed according to:

$$\phi' = \log_{10}K + 2\log_{10}L_{\infty}$$

with  $K$  put on annual basis, and  $L_{\infty}$  referring to total length, in cm.

### **Multivariate Extension of the Gulland-and-Holt Plot**

#### MULTIVARIATE APPROACH

Plotting the data of a single experiment in form of a Gulland-and-Holt plot, we will have a single regression line with some variance around the line. If, for example, from factorial experiments we have many different treatments and plot these all in the same Gulland-and-Holt plot, we will have a large scatter of points. Essentially, there are as many growth curves as experiments hidden in the cloud of points and these could be described by numerous VBGF parameter combinations. If we calculated a single regression from this scattered plot this would result in a pair of growth parameters expressing the central tendency in the whole of the combined experiments. Growth performance is governed by the concert of environmental variables encountered by the fish. The variance above and below the regression line can be attributed to the different factors or experimental treatments. The aim is to identify these variables, their interplay and to quantify their effects on growth, since they govern the shape of the growth curve (and the value of the corresponding VBGF parameters).

To explain more of the variance around the regression line and when numerous factors are involved, the bivariate regression can be extended to a multiple linear regression of the form:

$$y = a + b_1X_1 + b_2X_2 + \dots + b_nX_n$$

Thus, the Gulland-and-Holt plot can be extended into a multiple regression form (Pauly and Ingles 1981; Pauly and Hopkins 1983; Hopkins et al. 1988), permitting environmental and treatment variables to be considered simultaneously in the same analysis:

$$(L_2 - L_1)/(t_2 - t_1) = a + b_1\bar{L} + b_2X_2 + \dots + b_nX_n \quad \dots 11)$$

where

$$\bar{L} = (L_1 + L_2)/2 \quad \dots 12)$$

and where  $X_2 \dots X_n$  are environmental and treatment variables simultaneously recorded during the growth increments.

The idea is to relate all environmental effects (controlled treatments and uncontrolled variables) to the mean growth of fish during the investigated interval. Fish growth is used as an instrument to assess the effects of external variables, their direction (positive or negative) and their strength.

This requires that additional variables are measured simultaneously with fish size data. The VBGF parameters are estimated from:

$$K = -b_1 \quad \dots 13)$$

where  $b_1$  is the multiple regression coefficient of mean length. The value of  $L_{\infty}$  is the intercept of the multidimensional surface with the  $X_1$ -axis:

$$L_{\infty} = (a + b_2X_2 + \dots + b_nX_n)/-b_1 \quad \dots 14)$$

The information on the influence of environmental and treatment factors is now 'imprinted' in  $L_{\infty}$  and  $K$ .

#### CONFIDENCE LIMITS FOR $K$

The confidence limits for  $K$  derived with the multiple regression version of the Gulland-and-Holt plot are derived in the same way as for the simple Gulland-and-Holt plot, but with degrees of freedom =  $n-u$ , where  $n$  is the number of cases in the regression, and  $u$  is the sum of dependent and independent variables included in the regression.

#### CONFIDENCE LIMITS FOR $L_{\infty}$

Since  $L_{\infty}$  is obtained from a combination of the intercept, slopes and actual data values, the confidence limits are approximated using the method suggested by Sparre et al. (1989), described earlier in this paper.

### Length-Weight Relationships

In cases where only the weight of fish was taken, these values may be transformed to lengths with a length-weight relationship.

The relationship between length and weight of a fish species is generally described by the relationship (Ricker 1975; Pauly 1984):

$$W = v \cdot L^m \quad \dots 15)$$

or in a linearized form:

$$\log W = \log v + m \cdot \log L \quad \dots 16)$$

where

- W = weight of individual fish (individual values should cover a large range of weights)
- L = length of individual fish (individual values should cover a large range of lengths)
- v = intercept with ordinate
- m = exponent of length-weight relationship

The logarithms used are usually based 10, but base e can also be used. The exponent m usually varies around three (Carlander 1969; Pauly 1984). When the exponent is exactly three, growth is called "isometric", otherwise it is called "allometric". The coefficients describing the relationship of weight to length vary according to biological and environmental factors (Ricker 1975). For each stock or population under investigation the length-weight relationship should be determined separately over a size range as wide as possible.

We have not followed the suggestion of Ricker (1975) to use a "functional" (= Type II, or GM) regression for the length-weight relationship in this and other contributions in this volume and used instead a predictive (= Type I, or AM) regression.

Our reasons for using Type I relationships (Sokal and Rohlf 1969) are:

- we require length-weight relationships largely for predictive purpose;
- we consider W to vary more than L and measurement errors to affect W more strongly than L (the GM regression assumes X and Y to have similar variability and measurement errors);
- most software packages for statistical analysis incorporate Type I but not Type II regression routines; and

- when r is close to unity (as is generally the case when W and L values covering a wide range of sizes are related), the difference between the two regression types vanishes.

For conversions from weight to length, equation 15 can be rearranged to:

$$L = m \sqrt[m]{\frac{W}{v}} \quad \dots 17)$$

For the calculation of the allometric relationship (equation 15), the common practice is to linearize the variables through logarithmic transformation (equation 16) and to use linear regression to estimate the parameters v and m. This transformation introduces, however, a small systematic bias into the calculation when using the relationship for conversions. This can be accounted for with a correction factor (Finney 1941; Baskerville 1972; Beauchamp and Olson 1973; Whittaker and Marks 1975; Sprugel 1983), obtained from

$$F = e^{\left(\frac{SEE^2}{2}\right)} \quad \dots 18)$$

where SEE is the standard error of the estimate. It should be noted that this is based on natural logarithms, which should also be used in the regression. If the regression is determined on base-10 logarithms, this SEE should be transformed to natural base by multiplying the SEE with  $\ln(10) = 2.303$  before use in equation (18).

The correction procedure is to calculate predicted values with the allometric (here: length-weight) relationship and then to multiply each of these values with the correction factor (F) to eliminate log-transformation bias. Alternatively, the intercept v of the length-weight relationship can be multiplied with the correction factor (Vakily et al. 1986).

### Data Requirements

The method presented above relates growth increments over short time periods to environmental or treatment effects measured during and averaged for these time intervals (Table 1).

The data requirements therefore are:

1. A cultured population of aquatic organisms must be sampled in length and weight at regular, short intervals. For shorter intervals (e.g., less than two weeks at tropical

Table 1. Extended Gulland-and-Holt plot: data table organized according to experiment duration and individual measurements during intervals.

	Date	Length	Environment	
			Variable-1	Variable-2
Stocking	t1	L1	xi : xi	xi : xi
	$\Delta t$	L		
1st sampling	t2	L2	xi : xi	xi : xi
	$\Delta t$	L		
2nd sampling	t3	L3	xi : xi	xi : xi
	$\Delta t$	L		
.	.	.	.	.
.	.	.	.	.
.	.	.	.	.
Harvest	tn	Ln		

temperatures), growth in length will be difficult to detect and sampling stress may result. For longer intervals (e.g., six weeks or more at tropical temperatures), information will be lost. The sample sizes should cover a representative portion of the population. The data for each individual organism should be recorded.

2. All environmental and treatment variables of interest should be measured at regular intervals with the appropriate frequency to obtain representative values for these intervals.
3. In the design of factorial experiments for analysis by these methods, a wide range of values of each variable should be covered:
  - a) from small to large organisms, so that a representative growth model based on the VBGF can be fitted correctly and the model can be valid over a large size range of the species;
  - b) from low to high values of environmental and treatment variables, including an adequate number of zero-treatment (control) experiments, so that the regression can detect environmental and other effects on growth. Otherwise variables may become significant in the regression only due to natural or random variance; and

c) the range should be large in relation to the variance induced by the treatment variables.

4. Data must conform to basic assumptions of regression [see Hopkins et al. (1988) for a discussion of how these assumptions apply to the extended Gulland-and-Holt method].

### ***Computing the Extended Gulland-and-Holt Plot***

It is necessary to assemble all data in form of a data table for final multiple regression analysis. The first step is to tabulate all data according to sampling intervals (Table 1).


From the sampling intervals mean values are calculated for all variables during the interval, together with the time interval in days, the average length and the length increment. The data of all treatments and ponds are then organized in a data matrix ready to be used for multiple regression analysis. For the first pond and treatment the interval numbers are equal to the case numbers (Table 2). With this data matrix a multiple regression analysis is performed.

### ***Hardware and Software Requirements***

All steps of data compilation, processing and analysis can be performed on microcomputers. It is

Table 2. Extended Gulland-and-Holt plot: data matrix organized in final form appropriate for multiple regression analysis.

Case	Y	X1	X2	...	Xn
1	$\Delta L/\Delta t$	L	$\overline{\text{VAR1}}$	...	$\overline{\text{VARn}}$
2	"	"	"	...	"
3	"	"	"	...	"
.	"	"	"	...	"
.	"	"	"	...	"
n	"	"	"	...	"


  
 mean values of environmental variables in fish growth intervals

recommendable to employ spreadsheet software that is widely used (e.g., Lotus 123, Borland Quattro, Microsoft Excel) to construct several small spreadsheets of identical format for ease of data handling. These can later be combined for analysis. Several statistical packages for personal computers exist that perform multiple regression analysis and supply necessary statistics for regression diagnostics (e.g., SPSS/PC+, SAS, PC-Statistician, etc.). These all enable scientists in developing countries, without access to mainframe computers, to perform these analyses, and to exchange the data with colleagues for comparative analyses.

### Testing of the Model

The usual procedures and tests of statistical significance applied in least-squares regression should be applied. For a 'clean' hypothesis-testing approach (Prein et al. this vol.), each dataset was split into two parts by random sampling. One part contains 1/4 to 1/2 of the dataset and is used for model derivation; the other larger portion of the dataset is used for model testing (Norusis 1985).

Both portions of the datasets should produce regression models with the same set of significant variables. The signs and values of the regression coefficients should be the same, which can be tested by comparing the confidence intervals of the regression coefficients.

### Use of the Model

In determining the model with multiple regression analysis two goals can be achieved:

1. From a number of measured variables those with the strongest influence on fish growth should be identified. The rest will fall out as insignificant during analysis. Usually there are more insignificant variables than significant ones, leaving only few variables in the final equation.
2. The effects of the variables governing fish growth are quantified, each for itself and all together "in concert". The relative strengths of the variables can be compared.

With the Gulland-and-Holt plot, " $t_0$ " is not estimated since absolute ages are not known. Therefore a "recursive" form of the VBGF is used for predictions of growth (as originally published by von Bertalanffy in 1934):

$$L_2 = L_1 \cdot e^{-K\Delta t} + L_{\infty}(1 - e^{-K\Delta t}) \quad \dots 19$$

where  $L_1$  and  $L_2$  are the initial and final lengths, respectively, and  $\Delta t$  is the time interval of prediction.

With such a model the relative strengths of each of the variables can be used to deduce design and management implications in further culture operations. The whole model in itself can be used to make short- and long-term predictions of fish growth under certain, anticipated conditions (stocking density, feeding, temperature, etc.). Under given constraints, such as minimum market-size requirements or economic demands, the correct design and management scheme of a commercial culture operation may be determined using a biologically founded growth model incorporating environmental effects (Fig. 4).



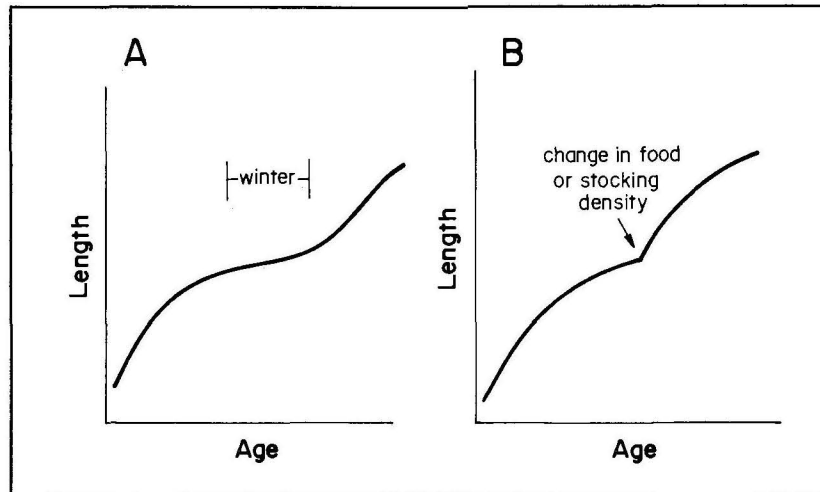


Fig. 4. Schematic examples for applications of the VBGF with inclusion of environmental and treatment variables to permit growth predictions under anticipated or planned conditions. A) High growth in summer and low growth in winter. B) Changes in feeding rate or stocking density can be modelled by the extended Gulland-and-Holt method.

Table 3. Some common problems encountered in aquaculture growth experiments and "solutions".

Problems	"Solutions"	Reference <sup>a</sup>
1. Changing environmental conditions during the culture period	a. Terminate the experiment when the change occurs b. Average growth over the whole period	Maguire and Hume (1982); Rappaport and Sarig (1979) Sin (1982)
2. Loss of a replicate due to circumstances unrelated to the experiment	a. Drop the replicate b. Use missing plot procedures	Hopkins and Cruz (1982) Gomez and Gomez (1976)
3. Limited number of ponds/culture units.	a. Limit the number of treatments and/or replicates. b. Use complex experimental designs such as fractional factorials, etc.	b Montgomery (1976)
4. Limited numbers of fish of one size for stocking	a. Multiple nursing b. Limit the number of treatments and/or replicates	Wohlfarth and Moav (1972) b
5. Changing growth responses as the fish grow larger	a. Conduct experiments over short size ranges only b. Average response over the whole size range	Knights (1983); Bryant and Matty (1981) Eldani and Primavera (1981)
6. Comparison of experiments which used different sizes of fish	a. Average growth rate using g/day or daily rates of increase, etc.	Coche (1982); Halevy (1979)
7. Estimation of size at time t	a. Use an average growth rate such as $g \cdot \text{day}^{-1}$ b. Use "sophisticated" growth equations such as von Bertalanffy, etc.	Huguenin and Rothwell (1979) Gates and Mueller (1975); Elliot (1975)

<sup>a</sup>Papers in which the "solution" is used or described.

<sup>b</sup>A standard technique which is usually not specifically mentioned by authors.

If mortality is negligible, fish yield at harvest is equal to the product of the number of fish stocked times the sum of measured growth increments. Alternatively, instantaneous mortality can be estimated (see paper by Hopkins and Pauly, this vol.) and included in the model.

### Discussion

Growout experiments play an essential role in aquaculture research in estimating the growth potential of various species and strains, in assessing the value of different feeds or treatments, etc. The problems with pond growout experiments are, however, that it is generally very difficult to control effectively the "control variables" and generally impossible to control extraneous variables (e.g., climatic factors) likely to affect the results of such experiments, resulting in "experiments" that in fact cannot be duplicated. Finally, since experiments are extremely costly in both time and resources, long-term aquaculture experiments that are intended to represent the time scale of commercial pond operations generally tend to be too limited to obtain secured and generalizable results (Table 3).

Explicit statements of this problem are few. Similarly, few papers are available in which optimal experimental designs for aquaculture research are discussed. This situation contrasts markedly with that prevailing in agriculture research, where experimental designs and analytical formats have traditionally benefitted each other, to the extent that whole chapters of statistical textbooks are devoted to them (Steel and Torrie 1960; Prowse 1968; Gropp 1979; Gomez and Gomez 1976).

In this paper, a method for the reduction and analysis of data from growout experiments was presented in form of the extended Gulland-and-Holt plot, which allows one to overcome the problems listed in Table 3. It is based on two assumptions whose validity can be assessed for any given set of experiments: a) that the growth of the fish can be described by the von Bertalanffy growth function, and b) that the effects of treatments and environment express themselves through changes in fish growth.

We conclude that the extended Gulland-and-Holt plot can be applied when the growth rate ( $dl/dt$ ) of the fish in the experiments can be described by the von Bertalanffy growth function (VBGF)

and when mainly environmental or treatment effects govern fish growth.

The applicability and benefit of this method for the analysis of aquaculture experiments has been demonstrated (Prein 1985, 1990, this vol.; Aquino-Nielsen et al., this vol.; Prein and Pauly, this vol.; Hopkins et al. 1988).

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