

On comparison of growth curves: How do we test whether growth rates differ?

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Comparisons of growth rates of populations and species are important in fisheries science for a range of reasons that vary with the context of each study. Most studies of fish growth have focused on the practical issues of the most appropriate way of comparing growth rather than on recognizing that there are several methods for making these comparisons and that the conclusions will differ depending on the method chosen.

Francis (1996) discussed the problem of how to compare different growth rates or growth curves. He suggested six plausible ways of making a comparison and suggested that the rate at which the asymptotic size is approached was the most natural method of comparing growth (his method 6). He illustrated the differences between the methods by comparing von Bertalanffy growth equations that are based on fixed growth parameters and that assumed that growth parameters are known and there are no associated uncertainties. However, in practice, the growth parameters are often estimated, and therefore are random variables. Consequently, the corresponding growth curves are also subject to variations.

For comparison with Francis's work, we will assume that growth for a spe-

cies is adequately described by the von Bertalanffy equation with $t_0 = 0$, as

$$L(t; \beta) = l_{\infty} (1 - e^{-kt}), \quad (1)$$

in which $\beta = (k, l_{\infty})$ are growth parameters.

Here $l(t)$ is the mean length at age t . If $\hat{\beta}_1$ and $\hat{\beta}_2$ are two estimates of β , how do we test whether the corresponding two growth curves are the same? The traditional way is to compare individual parameters and find out which ones are significantly different. However, the parameter estimates, l_{∞} and k , are often strongly correlated (Kirkwood and Somers, 1984; Wang and Thomas, 1995). It may therefore be more appropriate to compare biological reference points (e.g. size at one year of age) rather than growth parameters in the models (Wang and Thomas, 1995). Growth comparisons may, in general, be classified into two types: within species and between species. In practice, the following comparisons may be of interest:

- 1 Comparison of the growth rates for the same species, say E , in which two sets of growth parameter estimates, $\hat{\beta}_1$ and $\hat{\beta}_2$, are obtained from different time periods, different areas or sexes.

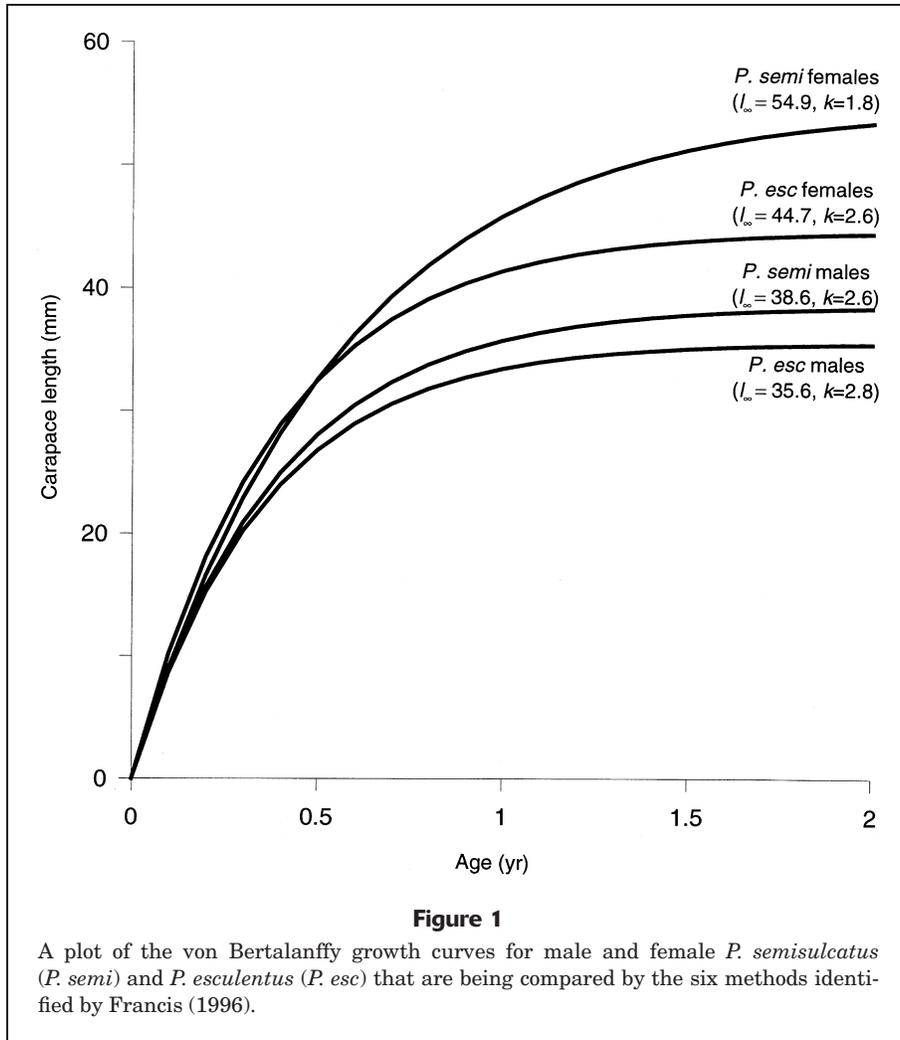
- 2 Comparison of growth rates for two different species to see which one grows faster.

As mentioned earlier, Francis (1996) considered six methods for comparing growth. For the within-species comparison, it seems all six methods are valid. However, these methods compare different characteristics of growth and therefore may reach different conclusions. For example, if we obtain $\hat{\beta}_1 = (0.5, 50)$ from area A and $\hat{\beta}_2 = (0.4, 60)$ from area B, we would conclude that species E does not grow as large in area A as in area B and that the k value (rate at which the asymptotic length is approached) in area A is larger than that in area B.

For between-species comparisons, we agree with Francis (1996) that his method 6 (k value comparison) is probably the most appropriate, especially in the context of comparing growth between, for example, herring and orange roughy. However, in some cases, comparing absolute growth rates at age or length between species could be of practical interest.

For example, if you are interested in choosing one of two species of fish or crustacean to farm and these two species look alike and have the same commercial value, it is more economical to farm the faster-growing species to shorten the time taken to reach market size. In Australia, the tiger prawn *P. esculentus* has a larger k value than the very similar *P. semisulcatus* (Somers and Kirkwood 1991), but *P. semisulcatus* has the potential of reaching a commercial size sooner (Fig. 1). Therefore, we would conclude that *P. semisulcatus* grows faster than *P. esculentus* in this context, and a comparison based only on k values may be misleading.

Therefore, in this note we will extend Francis's theoretical study by developing procedures for establishing statistical hypotheses for the six methods and suggest test statistics for comparing growth curves. We will demonstrate the differences in conclusions that can occur among the methods with data on



growth rates of two species of tiger prawn (shrimp) from northern Australia.

Methods

Overall hypothesis tests on two sets of parameters

Suppose we are interested in testing the hypothesis that the underlying growth curves corresponding to the two sets of parameter estimates $\hat{\beta}_1 = (k_1, l_{\infty 1})$ and $\hat{\beta}_2 = (k_2, l_{\infty 2})$ are the same. According to the large-sample theory, it is quite reasonable in most cases to assume that $\hat{\beta}_1$ and $\hat{\beta}_2$ are normally distributed. To be general, we will allow $\hat{\beta}_1$ to be correlated with $\hat{\beta}_2$. In notation,

$$\begin{pmatrix} \hat{\beta}_1 \\ \hat{\beta}_2 \end{pmatrix} \approx N \left(\begin{pmatrix} \beta_1 \\ \beta_2 \end{pmatrix}, \begin{pmatrix} \Sigma_1 & \Sigma_{12} \\ \Sigma_{12} & \Sigma_2 \end{pmatrix} \right) \tag{1}$$

Note that if $\hat{\beta}_1$ and $\hat{\beta}_2$ are estimated from different data sets, they may be assumed to be independent, because both

$\hat{\beta}_1$ and $\hat{\beta}_2$ are estimates and $\hat{\beta}_1 - \hat{\beta}_2$ is approximately multivariate normal. To test whether the two growth patterns determined by $\hat{\beta}_1$ and $\hat{\beta}_2$ are the same or not, we can use the generalised T^2 -statistic (Anderson, 1971):

$$T^2 = (\hat{\beta}_1 - \hat{\beta}_2)' V^{-1} (\hat{\beta}_1 - \hat{\beta}_2), \tag{2}$$

in which V = the covariance of $\hat{\beta}_1 - \hat{\beta}_2$.

The distribution of the T^2 -statistic is approximately chi-squared with 2 degrees of freedom, χ_2^2 . If the significance level is α , the corresponding critical value is $\chi_2^2(\alpha)$.

In many cases, we are interested in the slope of the growth curve (growth rate) rather than the curve itself. For example, we may be interested in comparing the growth rate during a particular age interval. Owing to natural mortality or fishing mortality, the period outside of this age range may be of no practical interest. In this case, it is may be more appropriate to consider the growth patterns over a specified age or length range rather than the whole range, which would put more emphasis (weight) on the asymptotic length in the comparison.

Tests to compare two growth equations Let g be a growth function of (k, l_∞) that we are interested in comparing. Table 1 lists the six g functions corresponding to the six methods identified by Francis (1996). For a given function g , we will test $g(\beta_1) = g(\beta_2)$ versus $g(\beta_1) \neq g(\beta_2)$ or $g(\beta_1) > g(\beta_2)$, depending on the context. Standard normal tests may be used for a specified g function. The test will rely on the properties of $D \approx g(\hat{\beta}_1) - g(\hat{\beta}_2)$. Let $E(D)$ and $V(D)$ be the corresponding expectation and variance of D when β is the true parameter. Under the null hypothesis, $g(\beta_1) = g(\beta_2)$, and using standard Taylor series expansion, we can work out analytic expressions of $E(D)$ and $V(D)$. Some pooled estimates of β may be required to input to $E(D)$ and $V(D)$ to obtain approximate values of $E(D)$ and $V(D)$. We can obtain $E(D)$ from $Eg(\hat{\beta}_1) - Eg(\hat{\beta}_2)$ and

$$E(g(\hat{\beta})) \approx g(\beta) + \frac{1}{2}(f_{11}\sigma_{11}^2 + 2f_{12}\sigma_{12} + f_{22}\sigma_{22}^2), \tag{3}$$

in which f values are from the second derivative of g with respect to β (Table 1) and

$$\begin{pmatrix} \sigma_{11}^2 & \sigma_{12} \\ \sigma_{12} & \sigma_{22}^2 \end{pmatrix} = V(\hat{\beta}). \tag{4}$$

The variance of D can be obtained from

$$V(D) \approx X_1^T \Sigma_1 X_1 + X_2^T \Sigma_2 X_2 - 2X_1^T \Sigma_{12} X_2, \tag{5}$$

in which X_i = the gradient or first derivative of g_i (Table 1); and

Σ 's = the components of the covariance defined earlier.

Note that the last term disappears if $\hat{\beta}_1$ and $\hat{\beta}_2$ are independent of each other. There are a few possible ways to obtain the approximate significance level, P . However, the most widely used method assumes D is normally distributed. Then we can use the z -test, which is based on the normal approximation for large sample sizes. For a one-sided test $g(\beta_1) = g(\beta_2)$ vs. $g(\beta_1) > g(\beta_2)$

$$P \approx 1 - \Phi \left(\frac{\hat{D}}{\sqrt{V(\hat{D})}} = \Phi \left(\frac{-\hat{D}}{\sqrt{V(\hat{D})}} \right) \right), \tag{6}$$

where Φ = the standard normal distribution function.

For a two-sided test, we have

$$P \approx 2\Phi \left(\frac{|\hat{D}|}{\sqrt{V(\hat{D})}} \right). \tag{7}$$

If we are interested in the growth for a range of ages (t_{min}, t_{max}), or the sizes (l_{min}, l_{max}), we may consider the in-

Table 1

The six growth models $g(\beta)$ that correspond to methods of comparing mean growth of two species or populations in Francis (1996) and their first $x = \frac{dg}{d\beta}$ and second $F = \frac{d^2g}{d\beta^2}$ derivatives assuming $\beta = (\beta_0, \Sigma)$.

Method	$g(\beta)$	X	F
1 Comparison of lengths at each age	$g = l_\infty (1 - e^{-kt})$	$\begin{pmatrix} l_\infty t e^{-kt} \\ 1 - e^{-kt} \end{pmatrix}$	$\begin{pmatrix} -l_\infty t^2 e^{-kt} & t e^{-kt} \\ t e^{-kt} & 0 \end{pmatrix}$
2 Comparison of absolute growth rates at each age	$g = l_\infty k e^{-kt}$	$\begin{pmatrix} l_\infty e^{-kt} (l - kt) \\ k e^{-kt} \end{pmatrix}$	$\begin{pmatrix} -(2 - kt) t l_\infty e^{-kt} & (1 - kt) e^{-kt} \\ (1 - kt) e^{-kt} & 0 \end{pmatrix}$
3 Comparison of absolute growth rates at each length	$g = (l_\infty - l)k$	$\begin{pmatrix} l_\infty - 1 \\ k \end{pmatrix}$	$\begin{pmatrix} 0 & 1 \\ 1 & 0 \end{pmatrix}$
4 Comparison of relative growth rates at each age	$g = \frac{k e^{-kt}}{1 - e^{-kt}}$	$\begin{pmatrix} \frac{1 - e^{-kt} - kt}{(1 - e^{-kt})^2} e^{-kt} \\ 0 \end{pmatrix}$	$\frac{kt^2(1 + e^{-kt})e^{-kt}}{(1 - e^{-kt})^3} - \frac{2te^{-kt}}{(1 - e^{-kt})^2} \begin{matrix} 0 \\ 0 \end{matrix}$
5 Comparison of relative growth rates at each length	$g = \frac{(l_\infty - l)k}{l}$	$\begin{pmatrix} \frac{l_\infty}{l} - 1 \\ \frac{k}{l} \end{pmatrix}$	$\begin{pmatrix} 0 & \frac{1}{l} \\ \frac{1}{l} & 0 \end{pmatrix}$
6 Comparison of rates at which the asymptotic size is approached	$g = k$	$\begin{pmatrix} 1 \\ 0 \end{pmatrix}$	$\begin{pmatrix} 0 & 0 \\ 0 & 0 \end{pmatrix}$

tegrated squared difference over the specified range. For example, if $g_1 = \lambda_1 e^{-k_1 t}$ and $g_2 = \lambda_2 e^{-k_2 t}$, in which $\lambda_1 = k_2 l_{\infty 1}$ and $\lambda_2 = k_2 l_{\infty 2}$, we will rely on

$$D = \int_{t_{\min}}^{t_{\max}} (g_1(t) - g_2(t)) dt.$$

The corresponding expectation and variance can also be approximated by the delta method.

We then apply each method to comparisons of the growth of males and females of two species of tiger prawn caught during tagging studies in northern Australia (Somers and Kirkwood 1991). We will consider two scenarios: 1) growth at age 0.5 yr (for methods 1, 2, and 4) or growth at length 30 mm (for methods 3 and 5); 2) growth at age one yr (for methods 1, 2, and 4) or growth at length 35 mm (for methods 3 and 5). In order to verify that our test assumption that D was normally distributed, we obtained frequency plots for the bootstrapped estimates of the growth parameters.

Results

We first bootstrapped the parameter estimates for each group to assess whether our assumption that the parameters were normally distributed was valid. The plot for male *P. semisulcatus* showed that there was little evidence of skewness in our estimates (Fig. 2). Plots for other groups are similar and not shown here. The t tests proposed in this paper, although based on normal distributions, are well known to be robust to violation to normality (which is why they are also known as “robust” test in statistics). On the other hand, the proposed tests rely only on mean and covariance estimates and the covariance matrix is often obtained from asymptotic distribution (normal) of the estimates in nonlinear regression.

We tested for the overall significance of each comparison before proceeding with testing the six methods. All were highly significant ($P < 0.00001$) and T^2 statistics ranged from 43.2 for the comparison of *P. semisulcatus* females versus *P. esculentus* females to 385.7 for *P. esculentus* males versus *P. semisulcatus* females. Given that the overall growth curves differed among species, then it is reasonable to then look further at the growth rates yielded by the different methods.

The growth models for the six methods of comparing growth and their first and second derivatives differed between methods (Table 1). The growth parameters used in the comparison show apparent differences in the size of both l_{∞} and k among the species and sexes (Wang, 1998). These differences in the absolute value of each parameter translate into quite large differences in the shape of the growth curves (Fig. 1). The effect is most striking for *Penaeus semisulcatus*, but does growth differ? Is it affected if we choose a particular length or age?

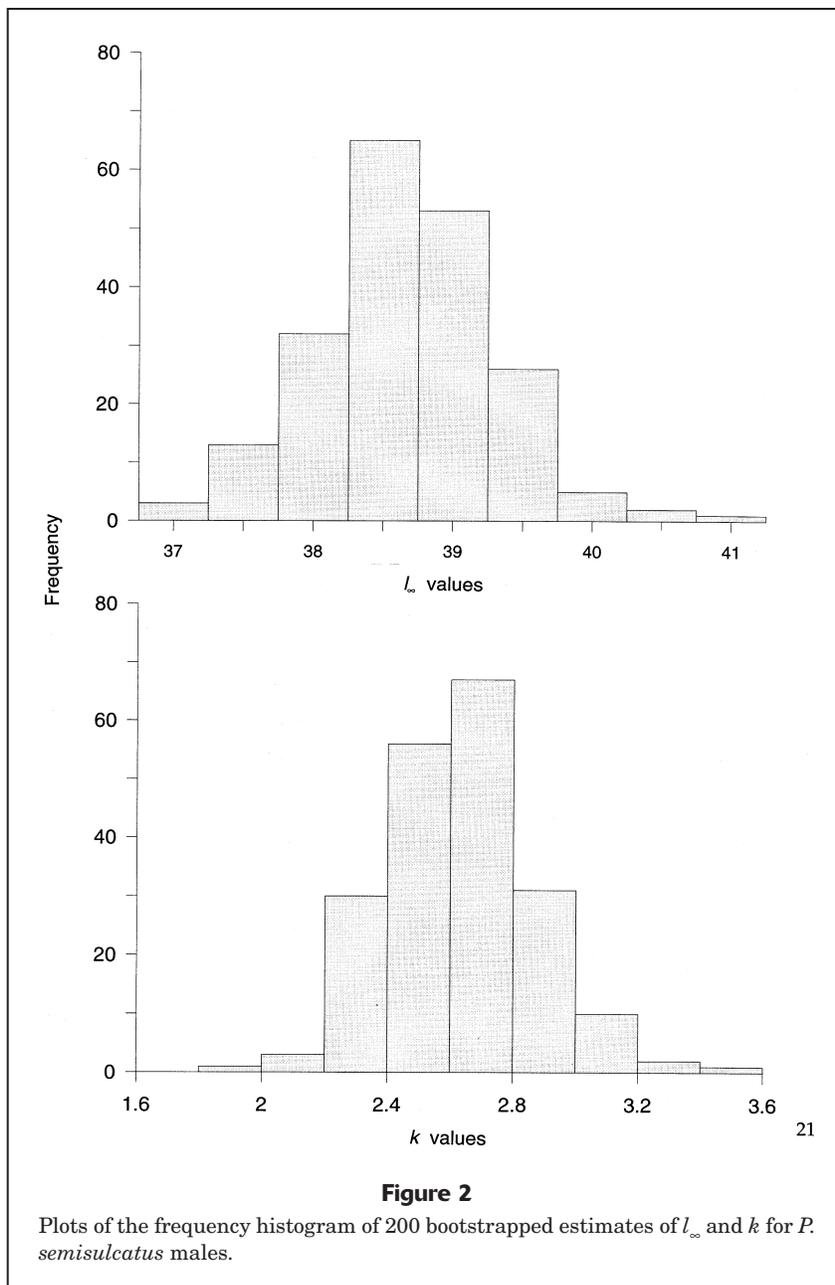
The results of tests from the six methods are shown in Table 2. The difference in lengths between *P. esculentus* and *P. semisulcatus* at the age of 0.5 yr is not significant for either males or females (in method 1 in Table 2), but the difference becomes very significant at age 1 yr. This result is consistent with the plot in Figure 1. However, the conclusion is reversed when comparing growth rate (method 2 in Table 2) instead of length. The relative growth rates at either age (0.5 yr or 1 yr) for *P. esculentus* do not differ significantly between males and females (method 4), whereas the rates at length 30 mm or 35 mm differ significantly. This is also biologically plausible because the growth rate at length 35 mm is very close to zero for *P. esculentus* males (the asymptotic size is only 35.6 mm) and there is a substantial growth to be gained for females (the asymptotic size is 44.7 mm). For method 6, the comparison is independent of specified length or age (two scenarios give the same results).

In prawn farms, *P. esculentus* and *P. semisulcatus* are harvested after 6 or 12 months, depending on the circumstances, such as the farmer’s need to increase the number of generations within the growing season at higher latitudes. Tests comparing the growth of each sex of each species (Table 3) show that the significance of the results varies with the hypothesis being tested. For example, the comparison of length-at-age (method 1) of *Penaeus semisulcatus* and *P. esculentus* and females was not significant ($P < 0.87$) at both six months of age (Table 3). However, when the absolute growth rates of the two species were compared (method 2), they were highly significantly different ($P < 0.001$). Other tests, such as comparisons of the growth of male and female *P. semisulcatus*, were highly significant for all methods ($P < 0.001$). As Francis (1996) pointed out, the results from all comparisons with methods 3 and 5 produce similar results and methods 4 and 6 give very similar results but in the opposite direction.

Table 2

The von Bertalanffy growth parameter estimates of *Penaeus esculentus* and *P. semisulcatus* from the Gulf of Carpentaria, northern Australia, used in growth comparisons between sexes of each species and between the two species for the same sex. The unbiased estimates were obtained by the equation of Wang (1998) and based on tagged prawn data from Somers and Kirkwood (1991).

Species	Sex	n	$l_{\infty} \pm SE$ (mm)	$k \pm SE$ (per yr)	Cov (l_{∞}, k)
<i>P. esculentus</i>	M	333	35.6 \pm 0.3	2.8 \pm 0.2	-0.0565
	F	224	44.7 \pm 1.2	2.6 \pm 0.3	-0.395
<i>P. semisulcatus</i>	M	159	38.6 \pm 0.5	2.6 \pm 0.2	-0.0886
	F	204	54.9 \pm 1.7	1.8 \pm 0.2	-0.2788



Our tests, in which the two tiger prawn species were compared, show that the significance of the results varies with the hypothesis. In our example, we have tried to choose lengths and ages that were approximately comparable. The results of the six tests for any particular interspecific or intraspecific hypothesis under either scenario are not directly comparable because methods 1, 2, and 4 compare growth rate at age (6 month or 1 yr) and the other methods compare growth rate in relation to length (30 mm or 35 mm). This distinction is more important for the interspecific comparisons because growth rate, rather than sexes, is more likely to vary between species for most phyla.

Discussion

Tests to compare growth by comparing length-at-age (method 1) show that there were no significant differences in the size of each sex of the two species, but there were highly significant differences in size between the sexes of each species. This finding differs from the interpretation if k -values were compared (method 6). In the situation where a farmer is deciding which species reaches a minimum marketable size earlier, we think that method 1 would be the most logical to use. However, if the farmer were trying to decide the potential benefit or tradeoff of

Table 3

Results of tests to compare within-species and between-species differences in the growth of males (M) and females (F) of two commercial penaeid prawns, *Penaeus esculentus* (*Pe*) and *P. semisulcatus* (*Ps*), from the Gulf of Carpentaria, northern Australia. The unbiased estimates from tagged prawns were obtained by the equation of Wang (1998). $D = g(\beta_1) - g(\beta_2)$, $V =$ the corresponding variance. Comparisons were made against the alternate hypothesis $g(\beta_1) \leq g(\beta_2)$ under two scenarios: 1) at age (t) = 0.5 yr for methods 1, 2, and 4 or at length (l) = 30 mm for methods 3 and 5; 2) at age (t) = 1 yr for methods 1, 2, and 4 or length (l) = 35 mm for methods 3 and 5. P_1 and P_2 are the P -values under scenarios 1 and 2.

Method	Comparison	Type of comparison	Scenario 1			Scenario 2			
			D	V	P_1	D	V	P_2	
1	Comparison of lengths at each age	within-species	<i>Ps</i> M vs. <i>Ps</i> F	-4.0	1.2	0.001	-9.7	0.4	0.001
			<i>Pe</i> M vs. <i>Pe</i> F	5.6	1.5	0.001	7.9	0.3	0.001
			<i>Pe</i> M vs. <i>Ps</i> M	1.3	1.0	0.18	2.4	0.3	0.001
			<i>Pe</i> F vs. <i>Ps</i> F	-0.26	1.7	0.87	4.2	0.4	0.001
2	Comparison of absolute growth rates at each age	within-species	<i>Ps</i> M vs. <i>Ps</i> F	-12.7	1.2	0.001	-9.2	2.0	0.001
			<i>Pe</i> M vs. <i>Pe</i> F	7.2	2.2	0.001	2.7	2.2	0.23
			<i>Pe</i> M vs. <i>Ps</i> M	2.8	1.3	0.03	1.4	1.3	0.29
			<i>Pe</i> F vs. <i>Ps</i> F	8.3	2.2	0.001	7.9	2.7	0.003
3	Comparison of absolute growth rates at each length	within-species	<i>Ps</i> M vs. <i>Ps</i> F	-21.4	1.6	0.001	-25.6	0.94	0.000
			<i>Pe</i> M vs. <i>Pe</i> F	22.3	2.3	0.001	23.4	1.08	0.000
			<i>Pe</i> M vs. <i>Ps</i> M	6.9	0.8	0.001	7.9	0.94	0.000
			<i>Ps</i> F vs. <i>Pe</i> F	5.9	2.7	0.03	10.03	1.09	0.000
4	Comparison of relative growth rates at each age	within-species	<i>Ps</i> M vs. <i>Ps</i> F	-0.27	0.08	0.001	-0.16	0.05	0.001
			<i>Pe</i> M vs. <i>Pe</i> F	0.06	0.11	0.59	0.03	0.06	0.60
			<i>Pe</i> M vs. <i>Ps</i> M	0.06	0.08	0.49	0.03	0.04	0.49
			<i>Ps</i> F vs. <i>Pe</i> F	0.27	0.12	0.03	0.15	0.06	0.02
5	Comparison of relative growth rates at each length	within-species	<i>Ps</i> M vs. <i>Ps</i> F	-0.71	0.05	0.001	-0.73	0.03	0.000
			<i>Pe</i> M vs. <i>Pe</i> F	0.74	0.08	0.001	0.67	0.03	0.000
			<i>Pe</i> M vs. <i>Ps</i> M	0.23	0.03	0.001	0.22	0.03	0.000
			<i>Ps</i> F vs. <i>Pe</i> F	0.20	0.09	0.03	0.29	0.03	0.000
6	Comparison of rates at which the asymptotic size is approached	within-species	<i>Ps</i> M vs. <i>Ps</i> F	0.84	0.26	0.001	0.84	0.26	0.001
			<i>Pe</i> M vs. <i>Pe</i> F	-0.22	0.4	0.58	-0.22	0.4	0.58
			<i>Pe</i> M vs. <i>Ps</i> M	-0.20	0.28	0.49	-0.20	0.28	0.49
			<i>Ps</i> F vs. <i>Pe</i> F	-0.82	0.39	0.03	-0.82	0.39	0.03

leaving the prawns in the ponds for longer, then one of the methods that test for relative changes in growth rate (method 4 or 5) would probably be more appropriate.

The difference in parameter estimates, and hence growth rates, may have important implications when researchers are trying to compare growth between species or between regions. Francis (1988) argued that length-at-age data do not contain precise information on the expected growth rate of fish of a given length. He concluded that length-at-age data and tagging data contain different information on growth; therefore the parameters estimated from them had different meanings and were not directly comparable.

Our proposed tests for different methods, including the overall test, aim to incorporate all the parameters and the overall uncertainties and correlations between each other. Further research can be carried out to study the robustness of the proposed test and to apply it to other growth models. The method suggested for constructing tests to compare growth rates could be easily modified to apply to other types of growth equation besides the von Bertalanffy equation.

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Literature cited

- Anderson, T. W.
1971. An introduction to multivariate statistical analysis. John Wiley & Sons, New York, NY, 284 p.
- Francis, R. I. C. C.
1988. Are growth parameters estimated from tagging and age-length data comparable? *Can. J. Fish. Aquat. Sci.* 45:936–942.
1996. Do herring grow faster than orange roughy? *Fish. Bull.* 94:783–786.
- Kirkwood, G. P., and I. F. Somers.
1984. Growth of the two species of tiger prawn, *Penaeus esculentus* and *Penaeus semisulcatus*, in the western Gulf of Carpentaria. *Aust. J. Mar. Freshwater Res.* 35:703–712.
- Somers, I. F., and G. P. Kirkwood.
1991. Population ecology of the grooved tiger prawn, *Penaeus semisulcatus*, in the north-western Gulf of Carpentaria, Australia: growth, movement, age structure and infestation by the bopyrid parasite *Epipenaeon ingens*. *Aust. J. Mar. Freshwater Res.* 42:349–467.
- Wang, Y.-G.
1998. An improved Fabens method for estimation of growth parameters in the von Bertalanffy model with individual asymptotes. *Can. J. Fish. Aquat. Sci.* 55:397–400.
- Wang, Y. -G., and M. R. Thomas.
1995. Accounting for the individual variability in the von Bertalanffy growth model. *Can. J. Fish. Aquat. Sci.* 52: 1368–1375.