Comments on the calculation of the specific growth rate in experiments with untagged individuals

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Summary: The specific growth rate, G, is widely used in articles dealing with the growth of aquatic organisms under experimental conditions. When individuals are untagged, the arithmetic mean of G for a group of animals must be calculated from weight geometric means, not from arithmetic means. The type of weight mean used in articles to calculate the arithmetic mean of G is usually not reported, and an extended use of weight arithmetic means is common. The arithmetic mean of G so calculated is biased according to the increment in the squared coefficient of variation of body weights. Another potential bias in the calculation of the arithmetic mean of G is size-dependent mortality; this bias cannot be avoided when individuals are untagged, but maximal and minimal values can be obtained. In summary, in view of these analytical results, it seems prudent to calculate the arithmetic mean of G for a group of untagged animals from geometric means of weights, and to estimate the maximal error due to the possibility of size-dependent mortality whenever possible.

Keywords: growth rate; corrections; aquatic organisms; experimental cultures; tagging; mortality.

Comentarios sobre el cálculo de la tasa específica de crecimiento en experimentos con individuos no marcados

Resumen: La tasa específica de crecimiento, G, es ampliamente utilizada en artículos que tratan sobre el crecimiento de organismos acuáticos bajo condiciones experimentales. Cuando los individuos no han sido marcados, la media aritmética de G para un grupo de animales debe ser calculada a partir de las medias geométricas del peso corporal, no a partir de las medias aritméticas del mismo. Habitualmente, en los artículos científicos no se explicita el tipo de media del peso que se ha utilizado para calcular la media aritmética de G, y el uso de la media aritmética de los pesos está extendido. La media aritmética de G así calculada está sesgada de acuerdo con el incremento en el coeficiente de variación cuadrático de los pesos corporales. Otro sesgo potencial en el cálculo de la media aritmética de G es la mortalidad dependiente del tamaño; este sesgo es difícilmente evitable cuando los individuos no han sido marcados, sin embargo se pueden obtener valores máximos y mínimos. En resumen, a la luz de estos resultados analíticos, parece prudente calcular la media aritmética de G para un grupo de animales no marcados a partir de las medias geométricas de los pesos, y estimar el error máximo debido a la posibilidad de mortalidad dependiente del tamaño siempre que sea posible.

Palabras clave: tasa de crecimiento; corrección; organismos acuáticos; cultivo experimental; marcaje; mortalidad.

The specific growth rate, G, is currently used to investigate growth patterns and growth correlates in both basic (Pelletier et al. 1995, Lefébure et al. 2011) and applied research (Fontagné et al. 2009) on aquatic organisms. The index is based on the concept of proportional growth and, for one individual, it can be

defined as the arithmetic mean of the instantaneous relative growth rate, dW/Wdt, over a finite time interval [0, t]. This simplicity trades-off against a number of dependencies on internal (body weight, genetic background) and environmental factors (temperature, diet) (Allen and Wootton 1982, Rabí and Maraví 1997, Björnsson and Steinarsson 2002, Lefébure et al. 2011). These dependencies make up the core of a number of papers dealing, for example, with fish growth: in basic research, the functional expressions connecting G and body size for a given species is frequently explored by means of correlation analyses in order to obtain a model for the growth trajectory (Björnsson and Steinarsson 2002); in applied research, G is widely used not to model growth, but to assess the performance of different treatments a posteriori (Fontagné et al. 2009, Collins et al. 2013).

Whatever the topic under investigation may be, it is necessary to calculate G according to its definition. In our view, if G is based on mean body weights, it should be based on geometric means of weight—otherwise the value worked out may be biased. The geometric mean for a numerical series is always lower than or as large as the arithmetic mean for the same series and the size of this difference depends approximately on the coefficient of variation for the considered set of numbers. Thus, a change in the coefficient of variation of fish weights over the experimental time can cause a bias in the calculation of G. The type of weight mean inserted in the formula of G is not always clear in scientific literature. It is often stated that averages are used, so an appraisal of the effect of inserting arithmetic means of weight in the formula of G seems to be prudent at the moment. In the case of experiments with tagged or individually reared animals, no bias exists if G is obtained from individual growth rates, but geometric means are necessary in the case of populations comprising untagged individuals reared in groups.

We would like to address the present comment to researchers mainly concerned with experimental designs on growth of aquatic organisms. No new growth index is proposed in this note. Instead, it is intended to briefly show our point of view about two types of biases potentially affecting the calculation of the well-known specific growth rate (biases that cannot be corrected during the statistical analysis of data) and to suggest a few recommendations in the calculation of this growth index, particularly in experimentation on juvenile animals kept in small groups. Let us now consider two situations of increasing complexity with untagged animals reared in the same population.

UNTAGGED FISH WITHOUT MORTALITY

Suppose a group of n untagged juvenile individuals in the same tank so that, as in many experiments with fish, n is not very high and body weights can be easily recorded for all animals at the first and the last samplings. In addition, suppose that there is no mortality. In such a simple experimental situation the arithmetic mean of individual specific growth rates, G, will be

$$G = \frac{1}{n}\sum_i G_i = \frac{1}{n}\sum_i \frac{1}{t}\left(\ln W_{i,t} - \ln W_{i,0}\right) =$$
$$= \frac{1}{t}\left(\frac{1}{n}\sum_i \ln W_{i,t} - \frac{1}{n}\sum_i \ln W_{i,0}\right)$$

Since the summation of the logarithms of i numbers equals the logarithm of the product of those i numbers and, in addition, the product of a constant by a logarithm can be written as the logarithm of a power expression, it is now possible to write,

$$G = \frac{1}{t}\left[\frac{1}{n}\ln\left(\prod_i W_{i,t}\right) - \frac{1}{n}\ln\left(\prod_i W_{i,0}\right)\right] =$$
$$= \frac{1}{t}\left[\ln\left(\sqrt[n]{\prod_i W_{i,t}}\right) - \ln\left(\sqrt[n]{\prod_i W_{i,0}}\right)\right]$$

By definition, the n-root of the product of i numbers is the geometric mean of those i numbers. Thus if μ_t and μ_0 denote the geometric means of the final and initial individual weights, the final expression of G becomes,

$$G = \frac{\ln\mu_t - \ln\mu_0}{t} = \frac{\ln\left(\mu_t/\mu_0\right)}{t}$$

The above expression is very similar to that for the specific growth rate for one individual, except that individual weights have been replaced by geometric means. If initial and final arithmetic means of body weights are used instead of geometric means, a bias is produced in the calculation. The quantity of this bias will depend on the change in the squared coefficient of variation for body weights, as explained below. The geometric mean, μ, can be approximated by developing the terms lnW_i, where W_i represents each individual weight, as a Taylor series centred at the value M, where M is the arithmetic mean of individual weights; then taking expectations, $E[\;]$,

$$\ln\mu = \ln M + \sum_{i=1}^{\infty} \frac{(-1)^{i+1}}{i\, M^i} E\left[\left(W_i - M\right)^i\right]$$

The Taylor series will be developed till the third term to obtain an approximation based on the arithmetic mean and variance of body weights; keeping in mind that the second term is zero, the sought expression is (Jean and Helms 1983)

$$\ln\mu \approx \ln M - 0.5\frac{V_w}{M^2} = \ln M - 0.5\,C^2$$

In the above approximation, V_w denotes the variance in fish weights and C sets for the coefficient of variation of fish weights. Now, substituting the above approximation into the expression of the true mean G,

$$G = \frac{\ln\mu_t - \ln\mu_0}{t} \approx \frac{\ln M_t - \ln M_0 - 0.5\left(C_t^2 - C_0^2\right)}{t}$$
$$G_M \approx G + 0.5\frac{\Delta C^2}{t}$$

where G_M is the growth rate as affected by the bias due to the use of arithmetic means of fish weights. The following results are now apparent:

$$\Delta C = 0 \Rightarrow G_M = G$$
$$\Delta C > 0 \Rightarrow G_M > G$$
$$\Delta C < 0 \Rightarrow G_M < G$$

The relative bias, $100 \times (G_M - G)/G$, can be calculated as

$$100 \frac{G_M - G}{G} \approx 100 \frac{\Delta C^2}{2\ln\left(M_t/M_0\right) - \Delta C^2}$$

For example, Petursdottir (2002) performed a series of experiments on the growth of tagged individuals of the arctic charr *Salvelinus alpinus* (Linnaeus, 1758); the M_t/M_0 ratio was in the range 1.8-2.1, and the initial and final coefficients of variation of fish weights were approximately 6.6% and 33%; if the experiments had been conducted with untagged fish, the relative bias due to the arithmetic mean would have been in the range 7%-10%.

UNTAGGED FISH WITH MORTALITY

Let us now think of a more complex but also more frequent experimental situation with juvenile animals: a group of untagged fish in the same container, whose body weights are recorded from time 0 to time t and with mortality in the same time interval; in addition, suppose that geometric means are used instead of arithmetic ones, so the bias due to the arithmetic mean can be ruled out. In this case, there is still a potential bias in the calculation of G, because the first sampling includes both dead fish and fish surviving to time t. This fact was early recognized in the field of fisheries research (Ricker 1975) and later on, and perhaps inconspicuously, in the field of fish culture (Otterå 1992). Moreover, some authors have followed the "mortality of the smaller" principle (Folkvord and Otterå 1993) to correct this potential bias in G.

The size of the selective mortality bias can be expressed as a function of fish size in the surviving and dead populations. This goal can be attained by splitting the factors within $\ln \mu_0$ (i.e. within the geometric mean of fish weights at time 0) into two groups according to the survival or non-survival of each individual and then rewriting the expression of G,

$$\ln \mu_0 = \ln \sqrt[n]{\prod_i W_{i,0}} =$$

$$= \frac{1}{n} \ln \left(\prod_{j \in surviving} W_{j,0} \right) + \frac{1}{n} \ln \left(\prod_{k \in dead} W_{k,0} \right)$$

Let s and d be the number of surviving and dead animals, respectively, μ_s and μ_d the geometric mean weights at time 0 of the surviving and dead individuals in the interval $[0, t]$, respectively, and $m=d/n$ the proportional mortality. Then,

$$\ln \mu_0 = \frac{s}{s} \frac{1}{n} \ln \left(\prod_{j \in surviving} W_{j,0} \right) + \frac{d}{d} \frac{1}{n} \ln \left(\prod_{k \in dead} W_{k,0} \right)$$

$$\ln \mu_0 = (1-m) \ln \mu_s + m \ln \mu_d$$

Therefore, it is possible to obtain an analytical expression for the bias, G_m–G (where G_m denotes the mortality-biased growth rate) as a function m, μ_s and μ_d. Firstly, consider the expression for G calculated regardless of the mortality,

$$G_m = \frac{\ln \mu_t - \ln \mu_0}{t}$$

Secondly, substitute the expression of $\ln \mu_0$ in G_m,

$$G_m = \frac{\ln \mu_t - \ln \mu_s + m \left(\ln \mu_s - \ln \mu_d \right)}{t}$$

At last, after some rearrangements, the relationship between G_m and G becomes,

$$G_m = G + m \frac{\ln \mu_s - \ln \mu_d}{t}$$

It is now apparent that the potential bias is dependent on the quotient of geometric mean weights of surviving and dying subpopulations. To our knowledge, this is a new result. Under positive mortality, three statements can be derived from the above expression,

$$\mu_s = \mu_d \Rightarrow G_m = G$$
$$\mu_s > \mu_d \Rightarrow G_m > G$$
$$\mu_s < \mu_d \Rightarrow G_m < G$$

In practice, because the values of μ_s and μ_d remained unknown in experiments with untagged fish, the size of the selective mortality bias, G_m–G, cannot be precisely estimated. Nevertheless, in the case of a researcher interested in getting an approximation to the actual value of G and acquainted with the exact mortality in the experiment, it can be suggested to calculate a maximum G value assuming that only the largest animals at the initial time died during the interval $[0, t]$, and also a minimum G value under the assumption that only the smallest animals at the initial time died during $[0, t]$. Thus the following estimation for the arithmetic mean of G can be useful for such an experimenter,

$$G = \frac{G_{max} + G_{min}}{2}$$

Except for the introduction of geometric means, the above expression is an extension of the correction of Folkvord and Otterå (1993), but it would be more appropriate when the validity of the "mortality of the smaller" principle is not clear. If the researcher is also interested in evaluating the goodness of the experimental measurement, the length of the semi-interval can be proposed as the maximal value of the error:

$$Error = \pm \frac{G_{max} - G_{min}}{2}$$

LIMITATIONS TO THE PROPOSED CORRECTIONS

It should be noted that in experimental scenarios where it is not possible to sample the whole population, for example when working with larval stages, the uncertainty of the calculated specific growth rate is also affected by the sampling error. Therefore, the

uncertainty in G will also depend on the statistical distribution of the selected metric variable—more specifically, on the shape of the distribution of $\ln W$ or $\ln L$. On the other hand, when the mortality rate is above zero, the calculation of $G_{\max}$ and $G_{\min}$ will in addition require knowing the true distribution of the population. However, these complications are beyond the scope of this comment.

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