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The growth rates of four populations of *Artemia franciscana* (Anostraca: Artemiidae)

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Abstract: The growth rates of three populations of *Artemia franciscana* from México (one from Yavaros, Sonora and two from San José, Baja California) and that of the reference strain from San Francisco Bay, California, grown under the same laboratory conditions, were not statistically different, either considering total mean increase in length, or increase by stages. However, interpopulational differences were detected in the final length of some developmental stages. In addition, regressive changes of dimensions were noted in some populations at the time of stage change. These might be the result of inborn characteristics of each strain, causing each population of *Artemia franciscana* to respond, in physiological terms, in different ways to the same environmental conditions.

Key words: *Artemia franciscana*, growth rates, intraspecific differentiation.

An organism's response to changes of its environment consists in adjustments in several physiological processes, all of which affect growth. Thus, in geographically isolated populations of a single species, local environmental differences and variations affect organisms in such a way, that their biological responses tend to be adaptive and consistent with the processes of natural selection. In this context, different populations of the same species may respond differently to the same environmental conditions, which in turn might be reflected by their growth patterns.

The effects of several abiotic and biotic factors, such as temperature, salinity, and food concentration and quality on *Artemia* growth, have been the object of many studies (e.g. Mason 1963, Reeve 1963, Sick 1976, Nimura 1980, Brune and Anderson 1989). However, a comparison and global analysis of the results of these works is difficult because no studies have considered inter- or intraspecific differences, which are known to exist at different levels. As a result of its allopatric distribution, several intraspecific genotypic differences have been described for populations of *Artemia franciscana* Kellogg of different geographic origin (Abreu-Grobois 1983). These have also been found in populations inhabiting different, though geographically close, types of environment (Correa-Sandoval 1991), and in this case differences have also been

documented at the biometric (Correa-Sandoval and Bückle-Ramírez 1993), reproductive (Correa-Sandoval *et al.* 1993a) and biochemical (Correa-Sandoval *et al.* 1993b) levels.

The present study was designed to ascertain whether these populations differ also in growth rates, when maintained in the laboratory under similar conditions.

MATERIAL AND METHODS

Cysts of the reference population (R) were provided by the *Artemia* Reference Center in Ghent, Belgium (*A. franciscana* from San Francisco Bay, California, U.S.A.). The others were from México: those of the first (Y) from Yavaros, Sonora (26° 40' N; 109° 35' W), identified as *A. franciscana* by Abreu-Grobois (1983) and those of the other two (A and C) from two separate ponds located in Ejido San José, Baja California (29° 15' N; 114° 53' W), also pertaining to the same species (Correa-Sandoval and Bückle-Ramírez 1993, Correa-Sandoval *et al.* 1993a)

The cysts were decapsulated with the hypochlorite method (Anonymous 1988) and the nauplii were collected within four hours after hatching. Four acrylic boxes (11 x 9 x 9 cm; actual water volume 500 ml) with a 150 µm mesh Nitex bottom were suspended in 15 L aquaria, with airstones placed directly underneath to aid with free exchange of water with the surrounding medium. The experiment was in triplicate. Initially, each box contained 2 000 nauplii of one population, for a total of 8 000 organisms in each aquarium. The populations are highly crowded, which lengthens the time to attain adulthood in order to obtain plentifulness of data for growth rates.

Water changes were once daily (40 %), using 1 µm-filtered, UV irradiated seawater. Throughoutt he experiment salinity ranged between 30 and 32 g L^{-1} and mean water temperature was 20.5 °C, above the low end of the optimum range for growth of the San Francisco Bay *Artemia* population used as a control (Sorgeloos *et al.*, 1986); the photoperiod was 24:00 light and the dissolved oxygen was 7.4 ppm.

The diatom *Chaetoceros* sp. (Strain CH-X-1 of CICESE's collection; Voltolina-Lobina *et al.* 1991), was used as food. Cultures were kept in semi-continuous 15 and 300 L cultures in f/2 medium (Guillard and Ryther 1962). The rations were initially as in Tackaert *et al.* (1987) with minor modifications (Table 1) but were kept constant after day 17.

TABLE 1

Rations of the microalga Chaetoceros *sp. provided to the populations of* Artemia franciscana *according to their age*

Artemia age (day)	Daily ration (cells*10^3/*Artemia*)
1	150
2-4	300
5.6	450
7	600
8	750
9	1120
10.11	1140
12.13	1800
14.15	2160
16.17	2520
Subsequent days	2750

Thirty organisms per day were collected in each box (ninety from each population) and preserved in scintillation vials with a non-deforming preserving solution (Correa-Sandoval and Bückle-Ramírez 1993). All specimens were measured for total length at a dissecting microscope with a calibrated eyepiece. After 48 days the organisms reached their maximum size, and the experiment was discontinued.

After checking their normality and homogeneity of variance, the data of the replicates were compared by one-way ANOVA (α =0.05). Since no differences were found, replicates from each population were pooled and analyzed together, again by one-way ANOVA and, when necessary, by the "a posteriori" multiple range test (Statgraphics PC v. 4.0).

Comparisons between populations were made considering overall growth, from first nauplius to final adult, and growth by stages. The day of change from one stage to the next was when >50 per cent of the specimens sampled for each population showed the morphological characteristics of the next older stage (Correa-Sandoval 1991). The durations of the various stages were the same for all populations and were as follows: nauplius: day 0 to 3;

metanauplius: day 4 to 18; juvenile: day 19 to 30; adult: day 31 to 48.

RESULTS AND DISCUSSION

In general, the four populations showed similar trends in growth rate, which increased from nauplius to juvenile and declined for the adult stage (Table 2). Rates were not statistically different ($P \leq 0.95$), either considering total mean increase in size, or increase by stages.

TABLE 2

Average growth rates (μm/day) global and by stages in differnts populations of Artemia franciscana *(standard error in parenthesis)*

	R	A	C	Y
Global	183 (19)	159 (19)	163 (18)	173 (24)
Nauplius	149 (6)	95 (64)	107 (28)	138 (29)
Metanauplius	173 (37)	176 (31)	168 (29)	227 (45)
Juvenile	238 (32)	206 (40)	225 (25)	243 (30)
Adult	138 (36)	78 (39)	47 (31)	98 (48)

R = Artemia *Reference Center; A = San Jose "A"; C = San Jose "C; Y= Yavaros.*

However, some of the stages final sizes were significantly different: from an initial nauplius situation $R \geq A \geq C \geq Y$ with $R > Y$ (421 μm±1.41 μm; 415 μm ± 6.1 μm; 408 μm ± 2.1 μm and 404 μm ± 1.0 μm), the final nauplii were $R \geq Y \geq C=A$ with $R > C=A$ (Table 3).The rest of the intermediate stages of the Yavaros population were consistently the largest, differences being always significant. For the final adults however, Y was significantly smaller than R (Y=6.75 mm ± 0.08 mm vs. R=7.42 mm ± 0.07 mm), both values were higher than those of the two San Jose populations (A=6.34 mm ± 0.05 mm; C=6.47 mm ± 0.05 mm) and these were not statistically different (Table 3).

These values are at the low end or lower than the size range between 6.87 mm and 7.60 mm reported for this species by Nimura (1980), definitely outside that of 9.20-12.45 mm mentioned by Reeve (1963) and lower than the 9.0 mm given by Sick (1976) (although these authors do not mention the species they studied, or refer to *A. salina*, their populations may be presumed to be *A. franciscana*, given their N. American origin).

Differences exist between the final length of one stage and the initial of the next one (Table 3). This difference was mostly positive, indicating an increase in size parallel to stage changes (A: metanauplius to juvenile, and juvenile to adult; C and R: metanauplius to juvenile). In the case of Y however, stage changes consistently coincided with significant decreases in length and a difference of this type, though not as well marked, was also noted for C (juvenile to adult: Table 3). These decreases suggest a considerable energy expenditure for stage changes probably as a result of different inborn characteristics of each strain, responding in dissimilar ways to proved culture conditions.

It seems necessary to comment on the long duration of this experiment, during which adulthood was reached in about one month. In other experiments with lower population densities but equal conditions of temperature and food availability (calculated as in this case as number of particles available to each individual) all four strains reached sexual maturity in about two weeks, within the time range reported in the literature (Sorgeloos, 1980).

Since neither of these factors may be considered responsible for the slow growth rates of this experiment, we believe that we introduced a stress factor (overcrowding), to which our populations showed a similar response but with different degrees of intensity. This enhanced the interpopulational differences, which might have gone otherwise undetected.

TABLE 3

Multiple range analysis (P≤0.95) of the initial and final sizes of Artemia franciscana *populations for each stage of development*

	(a)	(b)	(c)	(d)	(e)	(f)	(g)	(h)	(i)	(j)	(k)	(l)	(m)	(n)	(o)
	A-NF C-NF A-MI Y-MI C-MI Y-NF	A-MI Y-MI C-MI Y-NF R-NF R-MI	C-MF	A-MF R-MF	A-JI	R-JI	C-JI Y-JI	Y-MF	A-JF	A-AI	C-AI Y-AI C-JF R-JF	C-JF R-JF R-AI	A-AF Y=JF C-AF	Y-AF	R-AF
$\bar{X}$ (mm)	0.65	0.69	3.02	3.14	3.54	3.58	3.69	3.83	5.61	5.74	6.15	6.20	6.39	6.75	7.42
S.E.	0.03	0.03	3.02	0.03	0.02	0.00	0.00	0.03	0.05	0.01	0.03	0.03	0.04	0.08	0.07

R = Artemia *Reference Center; A = San Jose "A"; C = San Jose "C'"; Y = Yavaros; NF = nauplius final; MI = Metanauplius initial; MF = metanauplius final; JI = Juvenile initial; JF = Juvenile final; AI = adult initial; AF = adult final.*

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RESUMEN

Las tasas de crecimiento de tres poblaciones de *Artemia franciscana* de México (una de Yavaros, Sonora y dos de San José, Baja California) y una de la Bahía de San Francisco (E.E.U.U.) empleada como referencia, mantenidas bajo las mismas condiciones de laboratorio, no fueron estadísticamente diferentes, ya sea considerando el incremento promedio de la longitud total o el incremento por estadios. Sin embargo, se detectaron diferencias interpoblacionales en la longitud final de algunos estadios de desarrollo. Además, se observaron cambios regresivos de las dimensiones en algunas poblaciones al tiempo de cambio de la anamorfosis. Estas diferencias pueden ser debidas a características específicas e innatas de cada población de *Artemia franciscana* que se expresan ante una misma condición ambiental, en diferentes respuestas fisiológicas.

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