## The survival of *Nausithoe aurea* Silveira & Morandini, 1997 (Cnidaria, Scyphozoa, Coronatae) under extended periods of starvation\*

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**ABSTRACT.** The results of quantitative and qualitative starvation experiments with *Nausithoe aurea* polyps in natural and artificial seawater were compared. For this study, Scyphistomae growing on stony coral debris in the São Sebastião Channel (Brazil) were sampled by SCUBA divers. The data obtained reinforce the hypothesis about rearing differences between natural and artificial seawater. Some polyps showed better results in natural seawater than those in artificial seawater, with the former but not the latter surviving until the end of the experiment (155 days). These results suggest that Coronatae polyps use dissolved organic matter, which contributes to the survival of *Nausithoe aurea* polyps. During the experiments, the reaction of individual scyphistoma to gentle stimulation with forceps in the middle part of the periderm tube was also observed.

Key words: starvation, Nausithoe aurea, artificial seawater, Scyphozoa, Atlantic Ocean.

# La sobrevivencia de *Nausithoe aurea* Silveira & Morandini, 1997 (Cnidaria, Scyphozoa, Coronatae) en periodos prolongados de ayuno\*

**RESUMEN.** Mediante resultados cuali y cuantitativos se comparan experimentos sin alimento de pólipos de *Nausithoe aurea* en agua de mar natural y artificial. Se utilizaron pólipos colectados sobre restos de corales en la región del canal de São Sebastião (Brasil) mediante buceo autónomo con SCUBA. Se comprobaron diferencias en el cultivo entre los métodos utilizados. Algunos pólipos cultivados en agua natural mostraron mejores condiciones que los cultivados en agua artificial. Los pólipos en agua natural llegaron hasta el final de los experimentos (155 días), mientras que los cultivados en agua artificial no sobrevivieron hasta el final. Estos resultados sugieren el uso de materia orgánica disuelta por pólipos de Coronatae, la cual contribuye a mantener vivos los pólipos de *N. aurea*. Durante los experimentos se observó la reacción de los escifistoma frente a estímulos efectuados en la parte media del peridermia.

Palabras clave: ayuno, Nausithoe aurea, agua del mar artificial, Scyphozoa, Océano Atlántico.

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### **INTRODUCTION**

Coronatae polyps can produce ephyrae and/or planuloids as a result of strobilation (Jarms, 1997; Silveira & Morandini, 1997; Silveira *et al.*, 2003) and an operculum, whether peridermal or simply a tissue cover, may be produced near the aperture tube during strobilation (Werner, 1970).

Published observations of Nausithoe aurea

showed that Scyphistomae remained alive within their blocked tubes for over 13 months and that *Atorella vanhoeffeni* polyp strains kept in cultures survived for almost 40 years under laboratory conditions (Silveira *et al.*, 2003). Morandini & Silveira (2001a, 2001b) induced strobilation of *N. aurea* scyphistomae by offering them a surplus of available food. Unpublished data assured that, in laboratories, this coronate species always strobilates following

<sup>\*</sup> Trabajo presentado en el XXV Congreso de Ciencias del Mar de Chile y XI Congreso Latinoamericano de Ciencias del Mar (COLACMAR), realizados en Viña del Mar, entre el 16 y 20 de mayo de 2005.

large food availability since the surplus of energy stored by the polyps results in strobilation if there is not an annual regulation pattern (*sensu* Werner, 1979).

In their natural habitats, many benthic invertebrates are expected to use dissolved organic matter (DOM) as an additional food source (Southward & Southward, 1972; Shick, 1991; Acuña & Zamponi, 1995). Some authors discuss the great importance of DOM in the marine environment for the maintenance of life processes (see more in Valiela, 1995). For some groups, DOM uptake is a known source of nutrition (Southward & Southward, 1972; Shick, 1991; Acuña & Zamponi, 1995). However, the maintenance of vital functions without a regular feeding regimen and in which the nutrition supply is provided by the seawater (DOM) has never been recorded.

Although the importance of food to animals is common sense, the consequences of starvation have never been investigated in Coronatae. Other studies show the physiological influence of starvation for many higher animals (Paulay *et al.*, 1985; Strathmann, 1985), but such studies are still rather scarce for cnidarians.

The aim of this work is to test the survival of *N. aurea* scyphistomae over 155 days of starvation. Parallel feeding/starvation experiments were used to establish and compare possible patterns that control

strobilation. The expected influence of DOM was inferred from rearing the polyps in natural and artificial seawater. To analyze the influence of starvation on vital and normal functions, additional behavioral observations were made regarding the way polyps react to stimuli (contraction of the column and tentacles). This tentative approach and method were used to infer nutritional requirements through comparative differences in quantifiable responses.

#### MATERIALS AND METHODS

*Nausithoe aurea* polyps were collected on 23 June 2002 in the São Sebastião Channel, SP, Brazil (23°41' to 23°53.5'S; 45°19' to 45°30'W), a typical site (Fig. 1), by SCUBA diving (8-9 m). For details of the sampling methods, see Jarms *et al.* (2002) and Silveira *et al.* (2003).

The polyps were cultured following Jarms *et al.* (2002), but fed once a week with recently hatched *Artemia franciscana* Kellogg, 1906 nauplii. Ten scyphistomae were individually isolated in plastic Petri dishes (5-6 cm diameter, 25 mL seawater) and maintained under constant temperature (22°C) without light. Five polyps starved from 12 July to 17 December 2002 in natural seawater (NSW) that was changed weekly. Another five polyps starved from

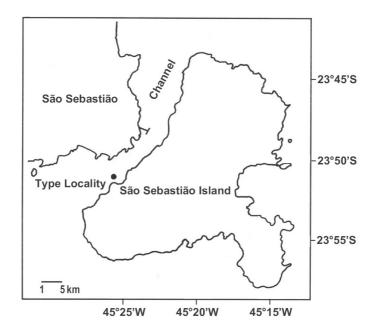


Figure 1. São Sebastião Channel (Type locality). Figura 1. Canal de São Sebastião (Localidad tipo).

29 December 2003 to 15 March 2004 in artificial seawater (ASW) (Tropic Marin®) changed weekly. Salinity and temperature were standardized, respectively, at 34-35 ppm and 22°C.

To compare the results of extended starvation, additional observations were made over 105 days (30 July-15 December 2002) with five polyps in NSW, starting and repeating 14-day starvation periods followed by 21-day feeding periods (five *Artemia* nauplii were offered each scyphistoma).

Additional observations of polyp behavior considered their reaction to stimuli: contracting the column and tentacles. Individual polyp responses were observed after a "touching test" (*sensu* Miglietta *et al.*, 2000) in which the middle part of the periderm tube was gently squeezed with sharp forceps to observe the possible retraction of the column and tentacles. The data were analyzed using the statistical program Minitab 14 and Excel for Windows.

#### RESULTS

During the experiment, no polyps starved in NSW were observed strobilating, but all those fed in NSW strobilated. No changes were observed in the polyps or in their responses to the touching test during the first 30 days of starvation. After this initial period, the polyps showed narrower columns, the gradual disappearance of the four mesenteries, diminished tentacle numbers, a thinner periderm tube, and a delayed response to the touching test. Tables 1 and 2 present the quantified observations of periderm tube growth for starved and fed polyps.

#### Starvation in ASW

No polyp strobilated during the experiments and the polyps reacted to the change from NSW to ASW by developing an operculum (Werner, 1970), which they shed after 2-6 days, and segmenting within the operculated tube (Silveira *et al.*, 2003). After 30 days, the response to touching or squeezing was very weak and two of the polyps reacted only after a second touch or squeeze. Table 3 shows the total length of the periderm tubes during the experiment. The starvation observations were concluded because all the Scyphistomae were in poor conditions by 15 March 2004.

#### **Touching test**

The reactions to the touching or squeezing test were standardized for all observed scyphistomae as partial withdrawal, total withdrawal, and contraction (Fig. 2). The responses were not uniform within the polyps in all groups. Some polyps failed to respond during the experiments (Fig. 3).

#### DISCUSSION

#### Statistical analysis

Simple statistics best illustrate the great changes observed in the normal conditions of the polyps during these experiments. The polyps in ASW (*i.e.* starved) decreased to an average of 0.179 mm, followed by the starved NSW polyps, which increased to an average of 0.186 mm; the fed NSW polyps increased to an average of 1.798 mm.



Figure 2. The progression of tentacle withdrawal and column contraction in *Nausithoe aurea* polyps. a) partial withdrawal, b) total withdrawal, c) contraction.

Figura 2. Progreso de la retracción de los tentáculos y contracción de la columna de pólipos de *Nausithoe aurea*. a) Retracción parcial, b) Retracción total, c) Contracción.

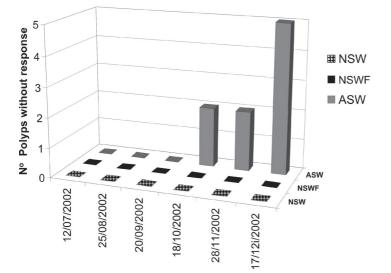


Figure 3. Results of touching test in *Nausithoe aurea* by experimental groups. NSW: natural sea water, NSWF: natural sea water fed, ASW: artificial sea water.

Figura 3. Resultados del test de estimulación en *Nausithoe aurea* en los grupos experimentales. NSW: agua de mar natural, NSWF: alimentados en agua de mar natural, ASW: agua de mar artificial.

Table 1. Total periderm tube length (mm) of starved *Nausithoe aurea* polyps in natural sea water. Note the reduction of the total length during the period (155 days).

Tabla 1. Longitud total (mm) de los tubos peridérmicos de pólipos en ayuno de *Nausithoe aurea* en agua de mar natural. Note la reducción de la longitud total durante el periodo (155 días).

Polyp	Α	В	С	D	E
July 12, 2002	3.80	4.00	5.20	4.80	3.85
October 03, 2002	4.40	4.03	5.20	4.92	5.16
November 28, 2002	3.62	3.90	5.23	4.80	5.30
December 17, 200	3.62	3.77	5.20	4.69	5.30
Amount reduced	0.18	0.23	0	0.11	increased 1.45

Table 2. Total periderm tube length (mm) of fed *Nausithoe aurea* polyps in natural sea water. Note the increase of the total length during the period (145 days).

Tabla 2. Longitud total (mm) de los tubos peridérmicos de pólipos alimentados de *Nausithoe aurea* en agua de mar natural. Note el crecimiento total durante el periodo (145 días).

Polyp	F	G	Н	Ι	J
July 30, 2002	2.60	2.70	3.40	4.40	2.30
October 07, 2002	3.00	3.24	4.90	4.90	3.12
November 17, 2002	3.50	3.71	7.52	4.99	3.23
December 25, 2002	3.90	4.01	8.04	5.02	3.42
Amount increased	1.30	1.31	4.64	0.62	1.12

Table 3. Total periderm tube length (mm) of starved *Nausithoe aurea* polyps kept in artificial sea water. Note the reduction of the total length during the period (113 days).

Tabla 3. Longitud total (mm) de los tubos peridérmicos de pólipos en ayuno de *Nausithoe aurea* en agua de mar artificial. Note la reducción de la longitud total durante el periodo (113 días).

Polyps	ASW1	ASW2	ASW3	ASW4	ASW5
December 12, 2003	4.200	4.225	6.225	3.475	3.425
January 01, 2004	4.200	4.226	6.225	3.476	3.420
February 11, 2004	4.123	3.936	6.145	3.235	3.419
March 15, 2004	4.063	3.936	6.095	3.142	3.419
Reduction amount	0.137	0.289	0.130	0.333	0.006

The distribution of the ASW data appears to be parametric; however, the NSW data were non-parametric. This is likely related to the polyps' probable nutritional uptake levels, which led to the following hypothesis: polyps in ASW (starved) cannot uptake the necessary food supply and variations in growth rates are impossible; polyps in NSW (starved) can uptake some food supply and variations in growth rates are possible; and polyps in NSW (fed) can uptake a surplus of food supply and variations in growth rates do occur. We believe that the groups reared in NSW will always present greater degrees of variation in their growth rates due to differences in their rates of nutritional supply uptake.

The results discussed above are clearly demonstrated in Figure 4; the data of all the polyps are combined in order to compare their growth rates. We observed an evident pattern within each experimental group: the starved polyps in NSW were mostly stable during the experiments (low growth rate), the fed polyps in NSW had the greatest growth rates (as expected), and the starved polyps in ASW had a considerably decreased growth rate.

#### **Biological implications for the Scyphistomae**

The fed scyphistomae had some sort of reserves within their tissues that enabled them to tolerate initial starvation (up to four weeks) without any damage

to their body structures (no observed modification in behavior or periderm tube structure). The polyps kept in NSW and starved remained in much better conditions over time than the starved polyps in ASW. Following Shick (1975), DOM (= alanine, glycine, and glucose-exposed) played an important role in maintaining strobilation and producing a number of ephyrae under prolonged starvation of Aurelia aurita polyps. Southward & Southward (1972) state that, although DOM uptake has not been proven essential for invertebrates, it might help them survive long periods of scarce food. Further evidence showing the likely importance of DOM in maintaining living tissue can be seen in the weakening of the periderm tube in the ASW experiment. Since most of the starved DOM-exposed polyps (NSW) also showed decreased periderm tube length, the secretion of the periderm seems to be controlled by a large food supply.

The fact that no polyp strobilated under prolonged starvation strongly suggests that energy storage and build up are required for the onset of strobilation in *Nausithoe aurea*. Some authors (Werner, 1973; Purcell *et al.*, 1999) affirmed that food storage is necessary for the onset of strobilation.

We suggest that the tube wall is maintained by active secretion within its inner surface as well as the addition of new material at the opening rim by the extension of the outer oral disc collar. No detailed in-

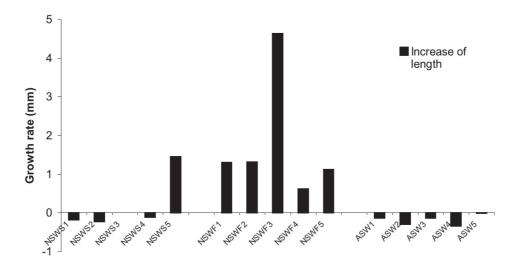


Figure 4. Combined growth rate (mm) data for *Nausithoe aurea* by experimental groups. NSWS: natural sea water starved, NSWF: natural sea water fed.

Figura 4. Datos combinados de la tasa de crecimiento (mm) de *Nausithoe aurea* comparados entre grupos experimentales. NSWS: ayuno en agua de mar natural, NSWF: alimentados en agua de mar natural. formation is available on the secreting activity or even the exact composition of the coronate tube wall.

#### ACKNOWLEDGMENTS

We thank Mrs. R.S. Morandini for Spanish comments, Mrs. V.B. Tronolone for the map, and Drs. A.C. Morandini and G. Jarms for helpful suggestions. The authors thank CEBIMar-USP for facilitating the specimen collection. SNS was supported by a Capes scholarship through the Graduate Program in Zoology, IBUSP. The Departamento de Zoologia, IBUSP, provided the continuous natural seawater supply. We are extremely grateful to two anonymous referees that made important comments and suggestions to this work.

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Recibido: 16 Mayo 2005; Aceptado: 16 Junio 2006

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