

FEEDING AND THE POSSIBLE ROLE OF THE PROBOSCIS AND
MUCUS COVER IN THE INGESTION OF MICROORGANISMS BY
RHYNCHOTEUTHION PARALARVAE (CEPHALOPODA:
OMMASTREPHIDAE)

Erica A. G. Vidal and Manuel Haimovici

ABSTRACT

The diets of 72 rhynchoteuthion paralarvae of *Illex argentinus* (Catellanos, 1960) (1.0-8.0 mm ML) and two other ommastrephid squids from southern Brazil (28°09'S-34°20'S) were investigated by examination of their digestive tracts and mucus covering. A great diversity of microorganisms was identified on the mucus cover, on the proboscis suckers and in the digestive tracts of the rhynchoteuthions, including dinoflagellates, flagellates, ciliates, cysts and bacteria. Among the digestive tracts of *Illex argentinus* rhynchoteuthions examined, 55.6% were empty, 9.7% contained unrecognizable food and 34.7% contained recognizable food, which included microorganisms on mucus as well as copepod appendages. Microorganisms on mucus were found mainly inside the digestive tracts of small paralarvae, which also displayed high bacterial densities on their mucus cover. The presence of bacteria on the mucus cover and of mucus in the digestive tracts decreased with increasing paralarval size. The smallest rhynchoteuthion with copepod appendages in its digestive tract was 3.7 mm ML. These findings suggest that mucus enriched with microorganisms may be important in the diet of small rhynchoteuthions, and it is hypothesized that mucus could act as a substrate for microbial growth. The proboscis may play an important role in the ingestion of mucus.

Recent reviews of the paralarval phase of cephalopods provide evidence that lack of food after absorption of yolk may be an important cause of mortality (Vecchione, 1987, 1991a), as has been shown for larval fishes (Hunter, 1984). Ommastrephid paralarvae are termed rhynchoteuthions because of the fusion of the tentacles into a proboscis. According to O'Dor et al. (1986), rhynchoteuthion paralarvae have a small quantity of yolk and a high metabolic rate, and may starve rapidly in the absence of food. High mortality rates calculated for *Todarodes pacificus* rhynchoteuthions were attributed mainly to starvation (Okutani and Watanabe, 1983). Available data suggest that mortality during the critical transition period after absorption of yolk to active prey capture is a common problem for all small-egg cephalopods (Boletzky and Hanlon, 1983; Balch et al., 1985; Vecchione, 1987, 1991a). This is particularly evident in all ommastrephid species, because all attempts to rear their paralarvae have failed. For *Illex illecebrosus*, it was suggested that the most obvious cause of such failure was the inability to induce feeding in newly hatched paralarvae (Balch et al., 1985). Until that time, no identifiable prey had been found within the digestive tract of wild-caught rhynchoteuthions (Durward et al., 1980; Balch et al., 1985, 1988; O'Dor et al., 1985). Lack of further information about the feeding of rhynchoteuthions may represent the major obstacle against keeping them in the laboratory. The development of techniques for culturing these commercially important squids will lead to an improved understanding of many aspects of their life cycles and behavior.

Based on the behavior of *I. illecebrosus* hatchlings during 9 d of survival in the laboratory, O'Dor et al. (1985) suggested that they could feed on suspended particles. Although rhynchoteuthions have no obvious sieving structures, they have large surface areas covered with mucus. These authors suggested that the paralarvae swim actively, and water flux on their outer surface is fast, and the body parts covered by mucus could retain particles by impaction. They also suggested that mucus is transferred from the mantle to the mouth area by ciliary motion and by cleaning behavior.

Vecchione (1991a) developed a method of clearing and staining cephalopod paralarvae, which makes the food particles in the gut easier to see; he found crustacean fragments in the digestive tracts of *Sthenoteuthis oualaniensis* rhynchoteuthions and the paralarvae of the enoploteuthid *Abralia trigonura*. The same method was used to examine paralarval feeding by *Lolliguncula brevis* (Vecchione 1991b).

Three rhynchoteuthion paralarval types are commonly found off southern Brazil: "types A", "B" and "C" described by Haimovici et al. (1995). "Type C" is *Illex argentinus* (Castellanos, 1960) and "type A" probably is *Ommastrephes bartramii* (Lesueur, 1821). The digestive tract contents of these rhynchoteuthion types, mainly *I. argentinus*, were examined in the present study. However, after detection of mucus in the digestive tracts, the mucus covering of paralarvae was also investigated to determine its role in feeding.

MATERIAL AND METHODS

All surveys were carried out between Cape of Santa Marta Grande (28°30'S) and Chui (34°20'S), Brazil, during spring of 1987, summer of 1990, autumn of 1991 and winter of 1988. The paralarvae were collected with a 60-cm mouth diameter, paired bongo net, with 0.33 mm mesh in oblique tows between the surface and approximately 300 m depth. Tows were carried out at a speed of 2 kt and deployed for 5-15 min, mainly during the day. The paralarvae were fixed and preserved in 4% buffered formalin.

Seventy-two digestive tracts of *I. argentinus* rhynchoteuthions from 1.0 to 8.0 mm mantle length (ML), 12 of rhynchoteuthion "type A" (1.0-5.8 mm ML) and four of the "type B" (2.0-4.0 mm ML) were examined. Paralarvae were stained with alcian blue and then cleared with trypsin following the method of Vecchione (1991a). Before the dissection of each paralarva, the external surface of the animal was examined to compare if particles attached to mucus were similar to those found inside the digestive tract. The stomach, caecum and intestine were then dissected and their contents analyzed with a light microscope at a magnification of 400× and 1000× when possible. The contents of the esophagus were not examined.

The digestive tracts examined (Fig. 1) were classified as: (1) empty; (2) with non-recognizable food: soft tissues in stomach and caecum, and digested food in the caecum and intestine; (3) with recognizable food: including crustacean fragments (mainly copepods), dinoflagellates, cysts, bacteria, flagellates, and ciliates.

Fourteen *I. argentinus* paralarvae (1.6-9.7 mm ML) were dissected with no previous clearing and staining treatment, but after fixation, to check whether the used method affects the identification of food items. The same food items were found in cleared and stained specimens and non-cleared and stained specimens, but in the untreated material, the opaque walls of the internal organs made the analysis more difficult. The strong reaction of mucus with the alcian blue made its visualization easy, both externally and inside the digestive tract. However, the mucus cover of paralarvae is very fragile and may be damaged or destroyed by handling or by the plankton net. Those paralarvae with the mucus cover partially destroyed were not examined.

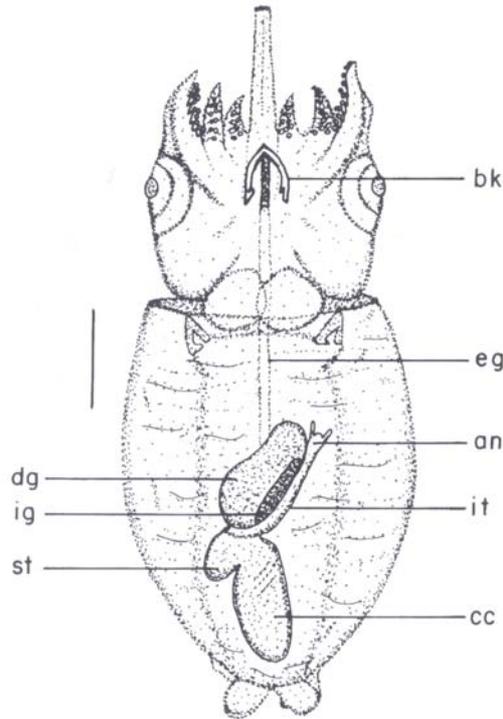


Figure 1. A schematic illustration of the digestive tract of rhynchoteuthion paralarvae; bk: beak; eg: esophagus; an: anus; dg: digestive gland; ig: ink sac; st: stomach; cc: caecum; it: intestine. Scale bar = 1mm.

Six of the treated *I. argentinus* digestive tracts had the stomach and/or caecum inflated with a dark fluid but contained no food. Since it is difficult to determine whether this fluid is a product of digestion (Vecchione 1991a), these digestive tracts were considered empty.

The presence of bacteria on the mucus cover was investigated in nine *I. argentinus* paralarvae (1.0-8.0 mm ML), five rhynchoteuthions "type A" (1.0-3.0 mm ML) and two rhynchoteuthions "type B" (2.0; 2.8 mm ML), previously cleared and stained. Five *I. argentinus* paralarvae (2.0-5.5 mm ML) we examined without previous treatment, but after fixation, to check whether clearing and staining caused bacterial contamination, or contributed to an underestimation of bacteria on the mucus cover. Pieces of paralarval mucus were gently removed, retained on a previously darkened Nuclepore filter (0.22 μm) and treated with the fluorochrome acridine orange (Hobbie et al., 1977). The filters were examined under an epifluorescence microscope (Axioplan Zeiss) using the filter sets 487709 (BP 450-490; FT510; LP 520), and a mercury lamp, HBO 50 W (Hobbie et al., 1977).

Two approaches were used to test the possibility that the high concentration of particles on the mucus of rhynchoteuthions was caused by the water flow in the plankton net. First, the density of bacteria and particles on rhynchoteuthions and on paralarvae of other cephalopods collected in the same tows, were compared, after clearing and staining, by light and epifluorescence microscopy. Secondly, the density of bacteria in the water remaining in the codend of the plankton net after tows of different duration was compared.

To estimate bacterial cell numbers, bacteria were counted in 30 to 60 randomly selected fields (1 μm^2 each) of the mucus of 13 rhynchoteuthions (1.0-8.0 mm ML) (considering types A, B and C and those treated and untreated), three loliginid paralarvae, *Loligo* spp. (1.7-3.0 mm ML), three enoploteuthid paralarvae, *Abralia* spp. (1.8-4.2 mm ML), and two chranchiid paralarvae, *Liocranchia reinhardtii* (3.8; 4.3 mm ML). The bacterial density μm^2 of mucus was then extrapolated to the total surface area of mantle. This area was determined

Table 1. Mucus cover examined under epifluorescence microscope and bacterial density found in treated and untreated rhynchoteuthions and other cephalopod paralarvae.

	Rhynchoteuthions		<i>Abralia</i> spp.	<i>Loligo</i> spp.	<i>Liocranchia</i> <i>reinhardtii</i>
Size range in mm ML	1.0-3.0	3.5-8.0	1.8-4.2	1.7-3.0	3.8-4.3
Number of mucus covers examined	14	7	3	3	2
Number of mucus covers suitable for counting bacteria	8	5	3	3	2
Bacterial density:					
close to zero	12%	40%	100%	100%	100%
low (<0.001 cells mm ^{sup-2})	25%	40%	-	-	-
high (>0.001 cells mm ^{sup-2})	63%	20%	-	-	-

considering the length of the mantle from which the mucus was removed and its diameter, assuming a cylindrical surface. Samples of water were collected from the water remaining in the codend of the plankton net after three consecutive oblique tows of 5, 10 and 15 min between the surface and 200 m in the study area. Immediately after the tows the watersamples were fixed with lugol. The enumeration of bacteria was also made by epifluorescence microscopy using the AODC technique (Hobbie et al., 1977).

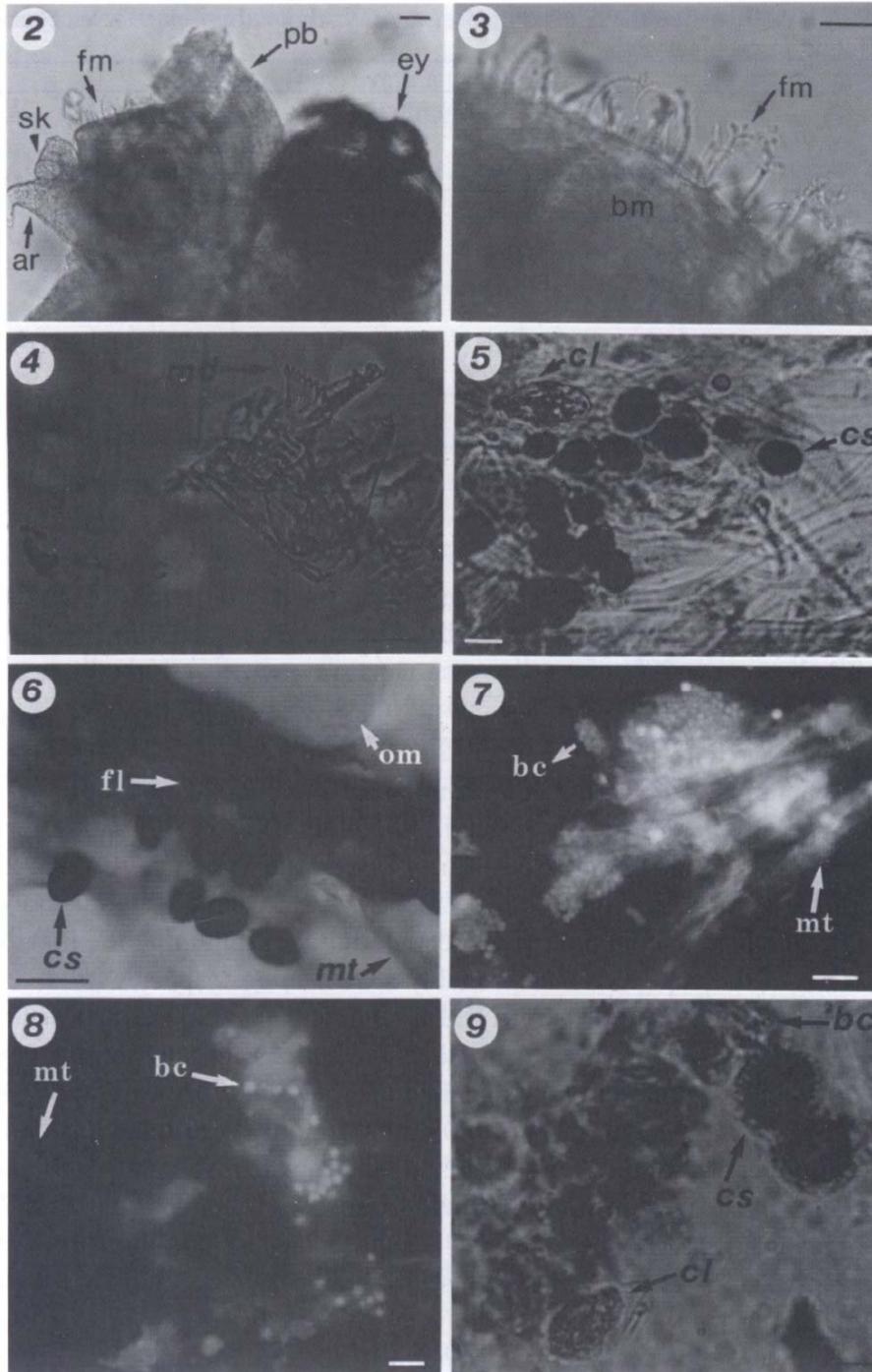
RESULTS

MUCUS AND MICROBIAL DIVERSITY. - A great diversity of microorganisms was observed in the mucus cover of all rhynchoteuthion types, including dinoflagellates, dinoflagellate cysts, coccolithophorids, autotrophic and heterotrophic microflagellates, ciliates, phytoplankton cells and very small bacteria-like cells (1 μ m), as well as crustacean fragments and detritus.

The acridine orange technique demonstrated the presence of high densities of cyanobacteria (1 μ m), heterotrophic bacteria (2-3 μ m), flagellates (2-4 μ m), ciliates and probably fungi (P. C. Abreu, pers. comm.), as well as other non-identifiable organisms on the mucus cover of those rhynchoteuthions treated by clearing and staining treatment and those untreated. However, the mucus of those untreated revealed the presence of other organisms, such as different ciliate and flagellate forms.

No bacteria were found on the mucus of *L. reinhardtii*. Only five to 20 small coccoid bacteria were found on the entire piece of mucus examined of *Loligo* spp. and *Abralia* spp., and none of the bacteria were associated with flagellates or ciliates. When these numbers were extrapolated to the total area of the mantle, bacterial density values were close to zero (Table 1). Rhynchoteuthions, however, presented considerably high values of bacterial density on the mucus, especially those from 1.0 to 3.0 mm ML (Table 1). Particles were also more numerous on rhynchoteuthions' mucus than in the mucus of the other paralarvae.

Rhynchoteuthions' mucus is relatively thicker than the mucus of *Abralia* spp., *Loligo* spp., and *L. reinhardtii*. Diverse particles were observed attached to the mucus cover of all rhynchoteuthions examined, although their density was variable. In general, the areas of highest concentration of particles were the posterior dorsal, ventral and lateral surfaces of the mantle, the lateral surface of the head, and the proboscis suckers. Sometimes mucus was found inside the funnel of some paralarvae. It was also observed that in rhynchoteuthions smaller than 2.0 mm ML, the lips of the buccal mass possess minute "filaments" (25-35 μ m). (Figs. 2,3), where accumulation of mucus and small particles (bacteria-like cells) were found. These "filaments" were only found in paralarvae up to 2.5 mm ML.



Figures 2-9: **2.** Head of a 1.0 mm ML rhynchoteuthion paralarva showing the localization of "filaments"; fm: "filaments"; ar: arm; sk: sucker; pb: proboscis; ey: eye. Scale bar = 60 μ m. **3.** Detail of the "filaments" of the rhynchoteuthion paralarva shown in Figure 2.; fm: "filaments"; bm: buccal mass. Scale bar = 20 μ m. **4.** Mandible of a 1.5-1.8 mm total length copepod and mucus found in the stomach of a 3.7 mm ML *Illex argentinus* paralarva; md: mandible; mc: mucus. Scale bar = 50 μ m. **5.** Microorganisms found in the mucus cover of a 1.5 mm ML *Illex argentinus* rhynchoteuthion; cs: "type 1" cysts; cl: ciliate. Scale bar = 10 μ m. **6.** Microorganisms found in the mucus cover of a 1.5 mm ML *Illex argentinus* rhynchoteuthion; cs: "type 1" cysts; fl: flagellates; om: organic material; mt: mucus thread. Scale bar = 10 μ m. **7.** Mucus cover of a 1.8 mm ML *Illex argentinus* rhynchoteuthion in epifluorescence microscope; bc, cyanobacteria; mt, mucus thread. Scale bar = 10 μ m. **8.** Mucus cover of a 1.5 mm ML *Illex argentinus* rhynchoteuthion in epifluorescence microscope; bc, heterotrophic bacteria; mt, mucus thread. Scale bar = 10 μ m. **9.** Stomach content of a 2.0 mm ML *Illex argentinus* paralarva; cs: "type 2" cysts; cl: ciliate; bc: probably bacteria on mucus. Scale bar = 10 μ m.

Table 2. Number of digestive tracts examined of the rhynchoteuthion paralarvae types and main food items found in each survey.

	<u>Spring 1987</u>		<u>Summer 1990</u>	<u>Autumn 1991</u>		<u>Winter 1988</u>
	<i>Illex</i> <i>argentinus</i>	type B	"type A"	<i>Illex</i> <i>argentinus</i>	type B	<i>Illex</i> <i>argentinus</i>
Number of stations with rhynchoteuthions	20	1	6	17	2	10
Number of stations analyzed	12	1	6	12	2	8
ML (mm) of examined rhynchoteuthions	1.0-8.0	2.8	1.0-5.8	1.3-5.5	2.0-4.0	1.0-7.8
Number of digestive tracts examined	41	1	12	19	3	12
Number of digestive tracts with copepods	5	0	1	2	0	2
Number of digestive tracts with microorganisms in mucus	8	0	2	5	0	3
Main prey	bacteria and copepods	-	-	"type 2" cysts	-	flagellates

The mean bacterial density from the samples of the codend of the three, different-duration tows were not significantly different based on ANOVA ($P > 0.05$).

FOOD AND FEEDING. - *Illex argentinus*. - Forty-one paralarvae from 12 stations were examined from the spring survey of 1987 (Table 2). A 3.7 mm ML paralarva contained fragments of two calanoid copepods (two cephalothoraxes including the insertion of the antennule). One of the copepods was probably a *Centropages* species (total length 1.5-1.8 mm). Fragments of its body, rostrum and antennules, mandibles, maxillae and maxillipeds were distributed throughout the stomach and caecum; the swimming legs were in the intestine. Mucus containing microflagellates, ciliates and bacteria was found in the stomach (Fig. 4). In two paralarvae (4.2; 8.0 mm ML), only the stomach contained body parts of a calanoid copepod; microorganisms in mucus were also found in the stomach of the smaller one, whose caecum was filled with fluid. Two other specimens (6.7; 7.3 mm ML) had copepod fragments in the stomach, but the caecum and intestine were empty. Two dinoflagellates (35-45 μm) of the genus *Prorocentrum* were found in the stomach of a 5.6 mm ML paralarva.

Cysts, probably of dinoflagellates, were found attached to the mantles and proboscises of 13 specimens collected at seven stations. These cysts (designated "type 1"), were elliptical, had a thick, dark rugose wall and measured 8-12 μm . In each of four specimens (1.5-3.0 mm ML) more than 1000 of these cysts were observed attached laterally and posteriorly to the mantle (Figs. 5,6). The cysts were associated with a complex microbial community, formed by cyanobacteria (Fig. 7), heterotrophic bacteria (Fig. 8), fungi, various flagellates, dinoflagellates, ciliates (Figs. 5,6) and coccolithophorids. However, "type 1" cysts were found in the stomachs of only two paralarvae (2.0; 2.8 mm ML), together with flagellates, ciliates and bacteria, attached to pieces of mucus. In some paralarvae, "type 1" cysts and heterotrophic bacteria were found on the "filaments" of the lips of the buccal mass. These cysts were not identified, but *Gymnodinium* spp., *Gyrodinium* spp. (>20 μm) and *Prorocentrum* spp. were found in high densities in surface phytoplankton samples (C. Odebrecht, pers. comm.) at the same stations where paralarvae had "type 1" cysts in the mucus.

Table 3. Contents of digestive tracts in different size classes of *Illex argentinus* rhynchoteuthions.

ML (mm)	n	Empty digestive tracts	Digestive tracts with non-recognizable food	Digestive tracts with recognizable food	
				Copepod fragments	microorganisms in mucus
1 - 2	20	13	2	0	5
2 - 4	31	18	3	1	9
4 - 8	21	9	2	8	2
Total	72	40	7	9	16
Percent		55.6	9.7	34.7	

Nineteen paralarvae from 12 stations of the autumn survey of 1991 were examined. In a 5.5 mm ML paralarva, the swimming legs of a copepod and two squid suckers were found in the terminal part of its intestine. Another paralarvae (4.7 mm ML) had a squid sucker and radula in its caecum. Five paralarvae contained mucus inside the gut. Three had "type 1" cysts on the mantle or proboscis, but in lower numbers than in the spring survey, and none were found inside the digestive tracts. Five paralarvae of this survey carried "type 2" cysts (probably dinoflagellate cysts) on their mantles. These cysts measured 15-20 μm , were spherical and had a thick wall covered with pointed protuberances (Fig. 9). In a 2.0 mm ML paralarva, three "type 2" cysts were found attached to the folds in the stomach wall (Fig. 9), and 11 more cysts, together with a high concentration of other particles, were observed on the mantle edge of a 3.0 mm ML paralarva, the head of which was withdrawn into the mantle.

Dinoflagellate cysts and high concentrations of dinoflagellates (*Dinophysis* spp., *Gyrodinium* spp., *Toridinium* sp. and *Proto-peridinium* spp.) were also found in surface phytoplankton samples (C. Odebrecht, pers. comm.) at stations where paralarvae had "type 2" cysts in the digestive tracts and on the mucus cover.

Twelve paralarvae from eight stations of the winter survey of 1988 were examined. A 4.7 mm ML rhynchoteuthion had numerous flagellates (4 μm) on the mantle and in the stomach, caecum and intestine. In a 3.8 mm ML paralarva, flagellates were found only in the stomach. A 5.0 mm ML specimen had crustacean swimming legs in its intestine, together with microorganisms in mucus. Another 7.8 mm ML paralarva also had hard parts of copepods in the caecum and intestine.

Feeding changes with growth of *I. argentinus* rhynchoteuthions. - Some evidence of changes in prey types of *I. argentinus* rhynchoteuthions with size was found (Table 3). In the smallest specimens the number of empty digestive tracts was high. Microorganisms on mucus inside the digestive tracts were found mainly in small paralarvae (e.g., 1.0-3.0 mm ML). Additionally, rhynchoteuthions of these small sizes had high bacterial densities on their mucus cover (Table 1). The presence of microbes on the mucus cover and of mucus in the digestive tracts decreased with increasing paralarval size. Mucus was not found in the digestive tracts of any paralarvae larger than 5.6 mm ML. Microorganisms on mucus, plus copepod fragments were found in the digestive tracts of paralarvae from 3.7 to 5.6 mm ML. The smallest paralarva with parts of copepods in its digestive tract was 3.7 mm ML. Only 12.5% of the digestive tracts contained hard parts of copepods, but the percentage of digestive tracts with copepods increased with size and was 38% in paralarvae >4.0 mm ML.

Rhynchoteuthion "type A". - All 12 "type A" rhynchoteuthions were collected during the summer. Only one paralarva (5.4 mm ML) contained copepod fragments in its digestive tract, while another (1.9 mm ML) contained non-recognizable food in the

intestine. Two other specimens (2.6; 3.2 mm ML) contained mucus. In one paralarva (3.6 mm ML), a mucus mass was found between the proboscis and the fourth pair of arms, and the external surface of the animal was relatively clean. Acridine orange treatment revealed a rich bacterial population on the mucus cover of these rhynchoteuthions.

Rhynchoteuthion "type B". - The four paralarvae examined were collected in the spring and autumn. All had empty digestive tracts, but their mucus covers contained microorganisms. Acridine orange treatment also revealed the presence of bacteria on the mucus.

DISCUSSION

The presented results are in general accord with the hypothesis of O'Dor et al. (1985) and provide evidence that early rhynchoteuthions may utilize enriched mucus as a food source. Most particles found on the mucus cover of rhynchoteuthions (e.g., bacteria, flagellates, ciliates, cysts) were also found inside their digestive tracts. Moreover, mucus may act as a substrate for microbial growth. This new hypothesis is based on two premises. First, microorganisms on mucus were found mainly inside the digestive tracts of small paralarvae, which also had high bacterial densities on their mucus cover. Second, bacteria were always found on the mucus of rhynchoteuthions associated with a microbial community. The ingestion of mucus with microorganisms could provide a means of obtaining energy before the animal attains the state of development necessary for capture of crustacean prey.

Durward et al. (1980) found high concentrations of protozoans colonizing non-viable eggs in *I. illecebrosus* egg masses in the laboratory. These authors suggested that in view of the limited swimming ability of rhynchoteuthions after hatching, egg masses may represent an adequate habitat for them, because of its available source of food. However, after leaving the egg mass, concentration of food would be much smaller.

O'Dor et al. (1985) suggested that rhynchoteuthions could concentrate particles through direct interception and inertial impact at low energetic costs with a surface covered with a viscous collector (mucus). They also suggested that after the impact of particles, mucus around the mantle could be transferred to the mantle edge by ciliary movements. The common procedure of withdrawing the head into the mantle, observed in live, post-hatchling rhynchoteuthions (O'Dor et al., 1985) and in preserved specimens (Okiyama, 1965; Haimovici et al., 1995), would be a more effective way of moving food particles from the head surface towards the mantle edge. The mantle edge would concentrate mucus from the mantle and head, making the ingestion easier. Some specimens with their heads withdrawn showed accumulations of mucus plus particles around the mantle edge, thus reinforcing this idea.

Mucus adhering to proboscis suckers was also frequently observed, indicating that the proboscis might transfer mucus from the mantle or head to the buccal area and so may play a role in the ingestion of mucus enriched with microorganisms. Movements of the proboscis towards the mouth were also reported by O'Dor et al. (1985).

The fusion of the tentacles into a unique proboscis-like structure may be related to such specialized feeding, especially in post-hatchling rhynchoteuthions that have rudimentary arms. In *I. argentinus* rhynchoteuthions, the proboscis begins to divide between 2.5-4.0 mm ML when the arms are relatively well developed (Vidal, 1994a). This may signal a change in feeding behavior. From 3.7 to 5.0 mm ML mucus and copepod fragments were found together in the digestive tracts. Vidal (1994a) suggested that the onset of active predation appears to be determined by the development of the arms and their suckers and not by the complete division of the proboscis as has been previously stated. The ingestion of enriched mucus could be particularly important for smaller rhynchoteuthions (e.g., <2.0 mm ML). Coincidentally,

rhynchoteuthions of these sizes possess "filaments" on the lips of the buccal mass (Figs. 2,3), where there are accumulations of mucus and bacteria-like cells. There are no previous records of the presence of these "filaments" on rhynchoteuthions. These "filaments" might represent an adaptation for the manipulation and ingestion of microorganisms or small pieces of enriched mucus. Further studies are necessary to investigate their structure and why they disappear in late paralarvae.

From the digestive tracts examined, only 12.5% contained hard parts of copepods. This number is similar to that obtained for *Sthenoteuthis oualaniensis* (Vecchione, 1991a), in which crustaceans were present in 16% of digestive tracts analyzed. In our study crustaceans occurred only in the digestive tracts of those paralarvae >3.7 mm ML. Rhynchoteuthions around this size already possess arm suckers in two rows, as well as on the proboscis tips, besides the eight previous suckers of 0.04-0.06 mm, several small suckers are being formed (Vidal, 1994a). All of these developments probably facilitate the seizure of copepods.

A great number of "type 1" cysts were found on the mucus cover of *I. argentinus* paralarvae, but were identified in the stomach of only two paralarvae, so it is difficult to determine whether they are a normal part of the diet. Cysts are resistant stages, and probably difficult to digest. Their high frequency might be due to their rough surface facilitating adhesion to the mucus cover.

This fact raises the possibility that the high concentrations of large particles, such as the cysts, found on the rhynchoteuthions' mucus could be caused by condensation in the cod-end of the plankton net. If so, this could explain the apparent negative selection of these cysts indicated by the gut contents.

The concentration of bacteria by the net was not observed. Additionally, very low numbers of bacteria were found in the mucus of the other cephalopod paralarvae collected in the same tows. The clearing and staining treatment also does not seem to influence the bacterial density, since high and low bacterial densities were found in both treated and untreated rhynchoteuthions. But, it is likely that condensation could happen to other relatively large particles like detritus and diatoms; the latter were sometimes found on the mucus cover, but not in the digestive tracts. Another possibility to be considered is that the presence of mucus in the digestive tracts could be a net-induced artifact. However, mucus was predominantly found in the gut of early paralarvae and was absent in larger ones.

The association of mucus with bacteria, flagellates and ciliates is well known and characteristic of a microbial aggregate ("marine snow", Alldredge and Silver, 1988). Appendicularian houses, pteropod webs and mucus released from coral reefs are common sources of macroaggregates (Davoll and Silver, 1986; Alldredge and Silver, 1988). Several authors (Sorokin, 1977; Linley and Newell, 1984; Biddanda, 1985, 1988; Biddanda and Pomeroy, 1988) have verified that bacteria are the first colonizers of mucus detritus of animal origin, followed by a succession of microflagellates and ciliates. The latter are consumers of bacteria, forming the microbial food chain of aggregates. This microbial heterotrophic community converts detrital material into living biomass and represents a potentially rich source of food.

Microbes could colonize rhynchoteuthions' mucus during locomotion or may be picked up from the egg mass after hatching. Phytoplankton and zooplankton detritus were commonly found on the mucus of rhynchoteuthions. This detritus may already be colonized by bacteria and even flagellates when it attached to the mucus, contributing to the development of the microbial populations on the mucus. Post-hatching paralarvae live in a low Reynolds number viscous environment, which may limit the potential for impact of particles because of the boundary-layer effects. However, only low collision probabilities would be necessary if mucus represents a substrate for microbes. Moreover, bacteria and flagellates have high production rates, doubling their numbers in a few hours (Brock and Madigan, 1991). It was

shown that reproductive efficiency of bacteria is even higher when they are attached to a surface (Alldredge et al., 1993). High concentrations of particles were found on the posterior and lateral mantle, areas probably difficult to reach with the proboscis. These areas could act as inoculum, in which bacteria could rapidly multiply and spread to newly secreted mucus.

Additionally, the feeding changes with growth found in *I. argentinus* rhynchoteuthions are in accordance with their distribution pattern off southern Brazil. Post-hatching rhynchoteuthions were found in areas of low zooplankton biomass and chlorophyll-a concentrations (Vidal, 1994b). The chlorophyll-a size classes comprised nanoplankton and picoplankton, which consisted of bacteria, phytoflagellates and small ciliates (mainly heterotrophic) i.e., the same microorganisms that were found on their mucus cover and gut.

Juvenile and adult cephalopods possess low digestibility of lipids and consume relatively small amounts of carbohydrates, but large amounts of protein, which is an energy source for them (Lee, 1994). Bacteria are largely protein. Thus one might expect that the enzymatic set that would be required for paralarvae to digest the microbial community found on the mucus could be the same one found in the adults. Recent histochemical studies of the digestive system of rhynchoteuthions revealed high proteasic activity (Boucaud-Camou and Roper, 1995). Phytoplankton, on the contrary, would require digestibility of cellulase, which was not found in rhynchoteuthions (Boucaud-Camou and Roper, 1995).

The consumption of microorganisms by rhynchoteuthions seems atypical for cephalopods, but it is common in other Mollusca. Pteropods (Thecosomata) secrete a mucous web many times larger than their bodies. A variety of microorganisms accumulate on these webs and are ingested (Lalli and Gilmer, 1989). Veliger larvae capture suspended particles by means of ciliary bands in the velum and secretion of mucus. Recent studies showed that some oyster larvae derive a large percentage of the total ingested particle volume from picoplankton size-particles (1-3 μm) and that these particles constitute a larger percentage of the material ingested by small larvae than that ingested by large larvae (Baldwin and Newell, 1995). Aggregation by mucus would help rhynchoteuthions to ingest very small cells like bacteria. The possibility of an association between microbes and rhynchoteuthions' mucus awaits additional investigations and represents an interesting topic for future studies.

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ADRESSES: (E.A.G.V.) *Marine Resource Center for Cephalopods, Marine Biomedical Institute, The University of Texas Medical Branch, 301 University Boulevard, Galveston, Texas 77555-1163*; (M.H.) *Departamento de Oceanografia. Universidade do Rio Grande, Rio Grande, Rio Grande do Sul 96201-900, Brazil.*