Water quality and zooplankton monitoring at the seawater supply circuit of a mollusc hatchery

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Seawater quality in mollusc hatcheries has been identified as one topic of high concern because of its implications in larval performance. This aspect, however, has received little or no attention from the scientific point of view. The only literature about this topic is a general compilation about water quality for aquaculture systems (Anonymous 1977). This gap in the available information may be mainly that periodic adjustments performed in hatcheries are routine protocols, the results of which are filed as internal reports and seldom reach the scientific literature.

Research done on this subject has dealt primarily with water quality and the composition of effluents from hatcheries and other aquaculture systems and their potential reuse for other purposes, such as agriculture and he culture of secondary aquatic organisms (Van Rijn 1996, Lin et al. 2002) There have also been studies associated with the effects of different treatments of seawater aimed at improving the quality of bivalve larvae (Utting et al. 1983, Chen 1984, Jaeckle and Manahan 1989).

Our study was triggered by the occasional finding of different types of plankton, mainly copepod microcrustaens, in the water used for larval culture in a mollusc hatchery. The assessment of water quality and biota throughout the entire water circuit was undertaken to improve the intake system and the treatment methods employed in conjunction with the water that circulates within the hatchery. This study is the first of its kind performed in Argentina.

We present here the results of a survey performed during a year in the hatchery during which water quality was monitored with regard to its physical and chemical properties, including temperature, salinity, pigments, organic matter. Planktonic species richness and total abundance, species abundance and the presence of non-planktonic organisms were also assessed.

Features of the Hatchery Seawater Supply System

The mollusc hatchery is located in the Northwestern coast of the San Matías gulf (40º48'S; 65º05'W; Northern Patagonia, Argentina). The building is placed on the dune line, at a distance of 150 m and a height of 18 m from the water line at normal high tide (tidal height = 7 m). A sandy beach is followed by a wide rocky intertidal zone of approximately 400 m. The hatchery has a lodging area, offices, one laboratory, an algal room, a big salon for broodstock conditioning and larval rearing and a larval culture room. An mechanical room is located next to the main building (Figures 1and 2).

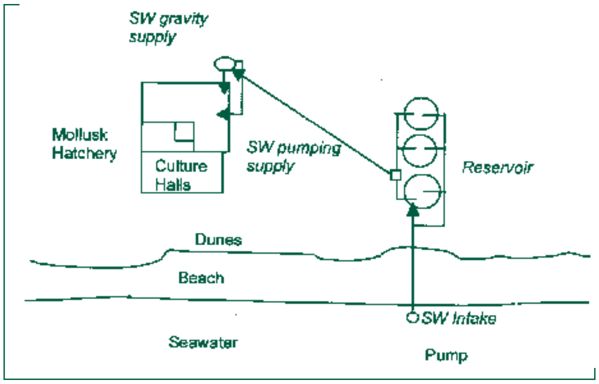

Fig. 1. Diagram of the seawater supply system of the mollusc hatchery.

Fig. 2. General View of the mollusc hatchery at Las Grutas beach nearly San Antonio Oeste City (Río Negro province, Argentina).

Seawater used in the hatchery is pumped during high tide from a square pool excavated in the upper rocky intertidal using a submersible pump (45 m^3/h) fixed to the wall of the pool. Water is stored in three, 43 m^3 concrete tanks located close to the hatchery.

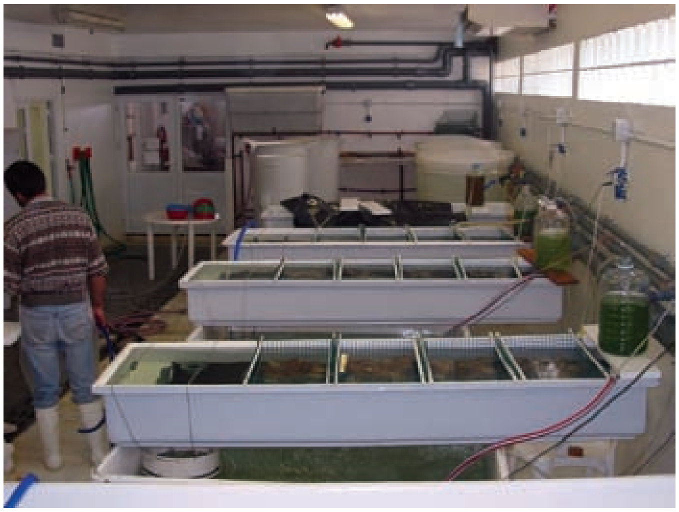

Fig. 3. Spawning hall.

Fig. 4. Algae culture hall.

Tank water is used 24 h after the tank is filled to allow sediments to settle. Water is then pumped into the mechanical room from which it is either forced into the hatchery by direct pumping, or pumped to an elevated tank (1000 L) from which it enters the hatchery by gravity. Two PVC pipes carry the water throughout the hatchery; one carrying unfiltered water used for broodstock and larvae and the other carrying filtered water used for algae and larvae. Pumped water is filtered through cartridge filters (10, 5 and 1 µm) placed in each room before being used for algal and larval cultures. Water used for algal cultures is additionally sterilized once in the culture bottles or polyethylene bags either by autoclave, for strains, or chlorination, for intermediate volume and mass cultures (Figures 3 and 4).

Sampling

Four sampling points (Figure 1) were selected along the water circuit: 1) intake, representing water in the sea during high tide, 2) reservoir, representing water existing within the external tank in use at that time, 3) gravity, water entering the hatchery by gravity from the elevated tank, and 4) pumping, water pumped into the hatchery. These four points were sampled bimonthly from May, 2000 to May, 2001.

At the intake point, water was sampled during high tide by pulling a zooplankton net (mesh diameter = 200 µm; mouth diameter = 0.30 m) by hand along a transect (100 m) at a speed of approximately 1 m/s. The mean volume of water filtered during each collection date was 14 m^3. The same procedure was used to collect a second sample with an 80 µm net to sample microplankton. However, the second sample was discarded after the first date because the composition of the sample was considered to be not representative of the water pumped into the hatchery. It contained low density detritus, fecal pellets, molts, algal pieces, and benthic organisms typical of the surf zone. At the remaining points, water was collected in buckets until volumes of 0.2 m^3 (reservoir) and 0.1 m^3 (gravity and pumping) were obtained. Water was filtered through a 80 µm net and concentrated material was fixed in formalin (neutralized 4 percent). Simultaneously, 1 L water samples were collected at each point to determine algal pigment chlorophyll *a* and phaeopigments, degradating chlorophyll *a*, particulate organic carbon (POC) and salinity. Water temperature was recorded at each sampling point. Total concentrations of chlorophyll *a,* phaeopigments and POC were analyzed according to Strickland and Parsons (1972).

Zooplankton was studied after fractioning each sample through a 200 µm mesh filter. Phytoplankton, detritus and organic sediment were also analyzed on the smaller size fraction. Quantitative and qualitative analyses were done in the conventional way using a stereoscopic microscope with appropriate chambers (Harris *et al.* 2000). Several aliquots of each concentrated sample were used for counts until reaching no less than 100 specimens per dominant species. Species classification was done using the available taxonomic literature (Pallares 1968, Lang 1975, Hoffmeyer 1983, Boltovskoy 1981, Boltovskoy 1999, Bradford-Grieve *et al.* 1999, Young 2002).

Temperature, salinity, chlorophyll *a*, phaeopigments, particulate organic carbon, species richness and zooplankton abundance data were statistically tested with a two-way ANOVA (sites and time, Table 1) to determine potential differences between sites and seasons.

Results

Seasonal and spatial behavior of abiotic and biotic variables

Temperature showed a seasonal pattern in agreement with natural coastal seawater behavior in the San Matías gulf, presenting a minimum in August of 9°C and a maximum in December of 17°C. In August, temperature at the intake was slightly higher than in the other points, while it was slightly lower during October, December and March; those

Table 1. Results of two-way ANOVA and Tukey Test. Data are for four sites and six sampling dates.

Test	Temperature (°C)			Salinity (ppt)			Chlrophyll a (µg/L)			Phaeopigments (µg/L)			POC[a] (µg/L)			Richness			Abundance		
	F	P[b]	S[c]	F	P	S	F	P	S	F	P	S	F	P	S	F	P	S	F	P	S
Tukey	32.6	0.00	**[d]	2.7	0.12	NS[e]	20.9	0.00	**	4.6	0.05	*[f]	1.2	0.29	NS	3.6	0.08	NS	24.9	<0.01	**
Site	5.7	0.01	*	3.5	0.04	*	0.8	0.50	NS	0.2	0.86	NS	1.0	0.44	NS	3.8	0.03	*	3.7	0.03	*
Date	280.2	0.00	**	129.1	0.00	**	3.6	0.02	NSD	1.9	0.15	NS	4.9	0.01	**	6.0	<0.01	**	4.3	0.01	**

[a] Particulate organic carbon
[b] Probability
[c] Significance
[d] Significant at $p \leq 0.01$
[e] Not significant
[f] Significant at $p \leq 0.05$

last two months showing the greater differences (Figure 5). Mean annual temperature on each site fluctuated between 13°C and 14°C, the lowest value corresponding to the intake. Temperature varied significantly among sites and dates (Table 1).

Salinity at all sites varied between a maximum of 34.64 psu (practical salinity unit) (October 2000) and a minimum of 31.77 psu (March 2001; Figure 5). Mean annual salinity observed in each site oscillated between 33.5 psu and 34 psu, with the highest value occurring in the reservoir. Significant differences were found between sites and dates.

Particulate organic carbon concentration at all sites showed a similar seasonal behavior than chlorophyll *a*, except for the peaks observed in May during both years (Figure 5). Mean values of all sites were similar, with the exception of pumped water which showed a lower concentration, while the highest seasonal variability was recorded in the reservoir. No significant differences were detected among sites, but highly significant differences were observed among dates.

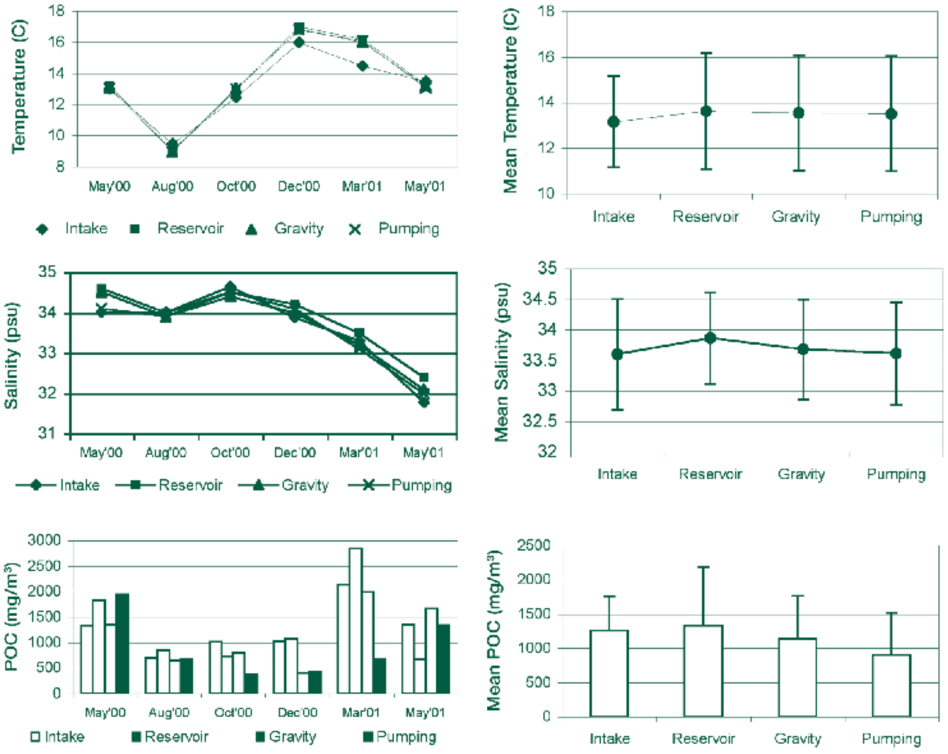

Fig. 5. Seasonal and spatial variability (among sites) in temperature (top), salinity (middle) and particulate organic carbon (POC, bottom). Values are expressed by site and sample date (left) and as mean annual values for each site (right).

The top of Figure 6 shows Chlorophyll *a* in surface water at the intake, which seasonally fluctuated from 4.67 µg/L, in March, to 0.05 µg/L in May. Water in external tanks (reservoir) and from internal taps (pumping and gravity) showed higher values during summer (1.73 and 3.13 µg/L, in December 2000 and March 2001). At those points values frequently were higher than at the beach intake (Table 2). The top right of Figure 6 details the mean annual values of Chlorophyll *a*, which were similar at the intake and in pumped water and higher than those in the reservoir and in gravity water. Seasonal variability in Chlorophyll *a* was higher at the intake, intermediate in pumped water, and lower in gravity fed water. No significant differences were found in Chlorophyll *a* between sites, while significant differences were found among dates.

The middle of Figure 6 shows that phaeopigment concentration behaved haphazardly without clear seasonality. During October, March and May the values were lower at the intake than in the tanks or inside the hatchery. Mean annual values were higher in the external sites (intake and reservoir)

Table 2. Composition and relative abundance (Nº ind/m^3) of zooplankton (>0.08 mm) at each sampling date and site. I: intake; R: reservoir; G: gravity and P: pumping.

DATE	05/29/2000				08/15/2000				10/25/2000				12/27/2000				03/10/2001				05/10/2001			
TAXA	I	R	G	P	I	R	G	P	I	R	G	P	I	R	G	P	I	R	G	P	I	R	G	P
Foraminifera	2	25	20	80	1											2								
Ostracoda		10											1											
Amphipoda	2	10		10	1								1										18	87
Oikopleura sp	1																							
Balanus sp (larvae)	3	5		20	5	5				200		6	1											
Bivalvia (larvae)		10		20			10					5		800		2,150								
Polychaeta (larvae)	2																							
Podon intermedius	11	5		10	4				25		1		1											
Evadne nordmanni								10																
Paracalanus parvus	10			20	1	10			17	200		8	40				7	133	150	111				
Ctenocalanus vanus					1	5							1	360										
Calanoides carinatus					1	5							1											
Labidocera fluviatilis										20												16		
Oithona nana	1		50	70					16	850		584	10	840		490	19		150	461	1			
Oithona similis	1								5															
Cyclopoida 1				10																				
Cyclopoida 2			10	10	2																			
Cyclopoida 3				10																				
Tisbe varians	1		10											1,300	5,450	520		133		223			18	95
Thalestris purpurea	1			10																				
Robertsonia propinqua	1																3							
Paralaophonte sp				10																				
Dactylopodia glacialis					4	5		30					1				3							
Orthopsyllus sp.													1											
Harpacticus gracilis															79									
Euterpina acutifrons												1												
Harpacticoida not id.				1									1											
Calanoida (nauplius)	1		10	10						683								515						
Harpacticoida (nauplius)				10																				
Euphausidae									2															
Fish (egg)									35				1											
Fish (larvae)									16				1											
Total abundance	37	65	100	301	20	30	10	40	116	1,953	1	604	61	3,300	5,529	3162	32	781	300	795	1	16	36	182

than in the water that reaches the hatchery, while seasonal fluctuations were higher in the reservoir than elsewhere. No significant differences were recorded between sites or dates.

The bottom of Figure 6 shows the variation of zooplankton total abundance and species richness. Zooplankton total abundance at the intake displayed its highest values in October 2000 (116 individuals/m^3), decreasing slightly thereafter. However, abundance increased dramatically in spring at the rest of the sites beginning with the reservoir in October and reaching maximal values at all sites during December (gravity water: 5,000/m^3; reservoir and pumed water: 3,000/m^3) . Mean abundance at the intake was much lower and showed lower seasonal fluctuations than at the other sites leading to statistical differences both between sites and dates. Species richness gradually decreased from May 2000 to May 2001. As a general rule species richness was higher at the intake than at the remaining sites, except during May of both years. Mean highest species richness was recorded at the intake and the higher seasonal fluctuations were observed at that site and pumped water. Species richness among sites and dates were significant.

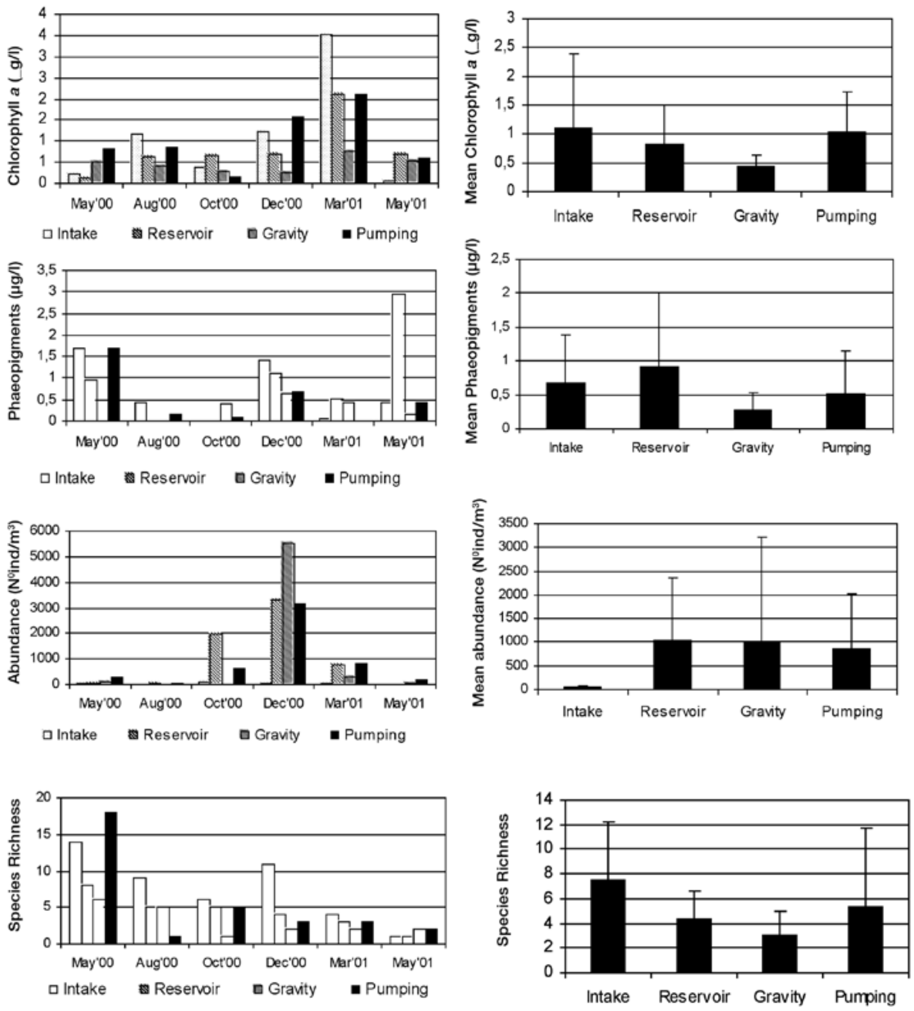

Fig. 6. Seasonal and spatial variability of chlorophyll a *and phaeopigments (top), zooplankton abundance and species richness (bottom). Values are expressed by site and sample date (left) and as mean annual values for each site (right).*

Zooplankton and other components

Material in samples from different sites was mainly composed of components larger than 200 µm while the smaller fraction, < 200 µm, only held some foraminifera and a few small nauplii of harpacticoid copepods.

The mesozooplankton, size greater than 200 µm, represented the following taxa: 18 species of copepods, four calanoids: *Paracalanus parvus, Ctenocalanus vanus, Calanoides carinatus* and *Labidocera fluviatilis;* five cyclopoids: three unidentified at the species level, *Oithona nana* and *O. similis* (=*O._helgolandica, sensu* Ramírez, 1966) and nine harpacticoids: *Tisbe varians, Thalestris purpurea, Robertsonia propinqua, Paralaophonte* sp*., Dactylopodia glacialis, Orthopsyllus* sp. and *Harpacticus gracilis, Euterpina acutifrons,* plus one unidentified species (Table 2). Nauplii of the three groups were also observed. Cladocerans were represented by *Podon intermedius* and *Evadne nordmanni*. Other organisms present were mollusc veliger larvae, nectochaete polychaete larvae and cirriped larvae. Occasional specimens of holoplankton and benthos included ostracods, amphipods and tunicates (*Oikopleura* sp.).

The most abundant species (number of individuals/m^3) at the intake from spring to fall were *O. nana, P. parvus* and *P. intermedius*. Those same species were found at that time in the reservoir, pumped water and gravity water, along with calanoid copepod and cirriped larvae and bivalve veligers. The harpacticoid copepod *T. varians* was almost not detectable at the intake even when it reached abundances of 1,300/m^3 in reservoir and 5,450/m^3 in the gravity water in December, 2000 (Table 2).

Planktonic algae were also observed (centric diatoms and *Ceratium* sp.) as well as molts of crustaceans; organic debris from invertebrates, other organisms and benthic algae; fecal pellets and sand grains. These components were more frequent in the intake, reservoir and gravity water than in pumped water.

Discussion and Conclusions

Statistical results obtained from the analysis of variance (ANOVA) performed on data from all the studied variables suggest a marked seasonal variability among dates for the variables, with the exception of phaeopigments. Also, a

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marked spatial variability among sites existed for all variables except chlorophyll *a*, phaeopigments and POC, which varied less.

Seasonal differences in intake water followed natural environmental fluctuations (Krepper and Bianchi 1982, Scasso and Piola 1986, Rivas and Beier 1990, Ramírez 1996). A similar pattern was found at the remaining sites with some variations resulting from the particular conditions and water management procedures used in each portion of the supply circuit. Fluctuations in water temperature in the reservoir water were usually in response to seasonal variations in air temperature. Salinity at the intake probably increased in summer because of evaporation resulting from high air temperature and heating of the incoming water running through the wide rocky intertidal zone.

Chlorophyll *a*, phaeopigments and POC varied among sites, with hatchery sites reaching much higher values than those recorded at the intake. The data suggest that from the reservoir on, and mainly in the elevated tank, several biological events were taking place: microalgal growth; degradation of chlorophyll into phaeopigments, resulting from consumption or algal alteration; consumption of herbivorous zooplankton; and accumulation of alochthonous material, fecal pellets and the remains of benthic algae, zooplankton and benthic animals.

Zooplankton species richness was higher at the intake than at the other sites. Conversely, zooplankton abundance showed a marked increase inside the hatchery, representing fewer species (*O. nana, T. varians, P. parvus* and cirriped, bivalves or copepod larvae). The behavior of these two variables confirmed the existence and active reproduction of populations of those species in the external tanks (reservoir) as well as in the elevated tank, more than in the remaining portions of the water supply system.

Zooplankton at the intake were comprised of species belonging to coastal-estuarine assemblages from temperate-cold latitudes commonly found in Argentina (Hoffmeyer 1983, Boltovskoy 1981, 1999). Seasonal succession agreed in general terms with available information from the San Matías gulf (Ramírez 1996), partly with that of Bahía Blanca estuary (Hoffmeyer 1994) and with the observations in Nueva Bay (Nuevo gulf) in Patagonia (Esteves *et al.* 1996).

Zooplankton species richness and total zooplankton abundance found in the intake water was less than that mentioned by Ramírez (1996) who reported 39 taxa, 15 belonging to copepods, from 36 stations within San Matías gulf (surveys onboard B/I Cruz del Sur, 1974-1975). In addition, while the copepods *O. helgolandica* (=*O. similis*), *P. parvus, A. tonsa* and *C. vanus* had been previously reported as the most abundant species in that gulf (Ramírez 1996), those species were present at low abundance during the present study. *O. nana*, mentioned as rare in the gulf by Ramírez (1996) was dominant in the seawater intake of the hatchery.

The lack of information corresponding to some months plus the variable conditions found in the water supply to the hatchery on different dates, caused by operation/managment differences, makes the interpretation of some results rather difficult. Nevertheless, and even when the results obtained show high seasonal and spatial variability, they provided important information regarding the efficiency of the filtration systems and the management protocols in use at the hatchery. Water treatment methods and management protocols were reformed based on these results and others suggested in the literature to improve water quality and biological production in this hatchery (Utting *et al.* 1983, 1985, Chen 1984, Jaeckle and Manahan 1989, Paillard *et al.* 2001, Renoult and Arzul 2001).

Notes

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