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ANIMAL SCIENCE

Evaluation of methods for reducing epibionts during farming of the mangrove oyster *Crassostrea gasar* (Adanson, 1757)

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Abstract: Due to the competition for food, space, oxygen and due to their role as diseases vector, epibionts can negatively affect oyster farming. We assessed the efficacy of six methods commonly used for the removal of epibionts from oyster shells during farming. The experiment was conducted at an oyster farm on the Paraná coast – South Brazil. Oysters (*Crassostrea gasar*) were acclimated for 90 d in the cultivation system and later exposed to cleaning treatments: i) freshwater; ii) hypersaline water; iii) sodium hypochlorite solution; iv) quaternary ammonia solution; v) exposure to air; vi) hydroblasting; and vii) no cleaning procedure (control). After treatment, oysters were kept in the cultivation system for 15 and 30 d – when the total incrustation and mortality were measured. Epibionts from nine phyla were identified. The most abundant were Arthropoda (Crustacea) (62.5%), Mollusca (33.8%) and Annelida (3.1%). Freshwater [15 (n = 2263 epibionts) and 30 days (n = 2822 epibionts)] and hydroblasting [15 (n = 1850 epibionts) and 30 days (n = 2389 epibionts)] treatments were the most efficient to reduce epibionts and caused lower rates of oyster mortality [15 (5.0 and 3.33%, respectively) and 30 days (1.67 and 6.67%, respectively)].

Key words: bivalve, commercial oyster cultivation, mariculture, oyster farming.

INTRODUCTION

During oyster farming, the structures used for cultivation (screens, ropes, floats, and Japanese lanterns), associated with the high density of oysters normally confined in a Japanese lantern, create an ideal condition for the establishment of other organisms (Adams et al. 2011); these organisms are known as biofouling or epibionts (Marshall & Dunham 2013). Ascidians, barnacles, hydroids, mytilids, and macroalgae are the most abundant epibionts found in cultivation systems of oysters farming (Ross et al. 2004). These animals are considered direct competitors as they vie for food, space, and oxygen (Arakawa 1990). The presence of epibionts can also increase the prevalence of diseases during farming (Mohammad 1976, Taylor et al. 1997, Guenther et al. 2006). Thus, inadequate management during farming could increase the presence of epibionts and adversely affect oyster growth and survival rates and meat yields (Pauley & Troutt 1988).

The presence of epibionts on oyster shell could also compromise the visual appearance of the marketed product (Doroudi 1996). For example, Enright (1993) and Claereboudt et al. (1994) estimate that the presence of epibionts on farmed oysters cause reduction of the product final price, which could result in losses from 5 to 30% of the worldwide shellfish production. Their presence on the cultivation system can also increase the weight of the structure, compromising durability, damaging the material and hinder the operational management (Fitridge et al. 2012). All these consequences may, in turn, affect the economic viability of oyster farming (Pit & Southgate 2003).

Due to the presence of epibionts during the farming process, oysters and cultivation structures must be periodically cleaned to reduce incrustation. Consequently, the cleaning methods and techniques need to be as simple, fast, efficient, safe (for the operator, the oysters, and the environment) and non-residual as possible. Currently, three main cleaning methods are currently adopted: mechanical/ physical control (manual removal method, hydroblasting, air exposure, exposure to high water temperatures) (Cheney 2010); chemical control (immersion in freshwater or hypersaline water, or immersion in sodium hypochlorite, quaternary ammonia, acetic acid, hydrated lime or sodium hydroxide solutions) (Piola et al. 2010, Rolheiser et al. 2012); and biological control [use of predators (urchin, Mediterranean acoglossan mollusks) of these epibionts in the structures or cultivations areas] (Mcennulty et al. 2001).

The goal of the present study was to evaluate the efficacy of six methods (immersion in fresh and hypersaline water, in sodium hypochlorite solution, in quaternary ammonium solution, hydroblasting and air exposure) normally used for removing.

MATERIALS AND METHODS

Farming area

The experimental procedure was conducted on a farm located at Rio dos Pinheiros, in Guaratuba Bay, Paraná, Brazil (25°50'02.34"S 48°34'45.40"W). The Guaratuba Bay is an estuarine area (48.57 km²) surrounded by marsh vegetation and mangrove forest. Cubatão and São João rivers are the main sources of freshwater into the Bay, with an average input of 80 m3/s (Marone et al. 2006). In some areas, water depth reaches 6 m and salinity, and temperature can vary from 3 to 37 g/L and 15 to 28 °C, respectively, influenced

by seasonal changes (Chaves & Bouchereau

1999, Brandini 2008). At this farm, the oysters (Crassostrea gasar) are typically cultivated in Japanese lanterns from juvenile phase (~2.0 cm) up till commercial size (7.0 to 8.0 cm). The Japanese lanterns (total height= 60 cm, diameter= 40 cm, number of floors= 4, height of each floor= 13 cm, mesh size= 3 cm) were suspended on floating long lines, at 1 m of distance from each other. In the final stage of farming (~ 8 cm in height) the density 105 utilized is 60 oysters (5 dozen) per floor.

Experimental design

The experiment was conducted during the fall season. During the acclimation period (90 d), oysters were kept in Japanese lanterns without cleaning managed. Before the beginning of the experiment, 60 oysters were randomly sampled for qualitative and quantitative characterization of the total of epibionts originally present on their shells and biometric measurements (time 0). Then, oysters (n= 840; mean weight= 70 g) were selected by size (7.0 to 8.0 cm) and divided in six groups (treatments), to which the different epibionts cleaning methods were tested (n=120/ treatment), and one control group (n=120) (Table I).

Cleaning methods

The tested methodologies were: immersion in freshwater, immersion in sodium hypochlorite solution [100 ppm], immersion in quaternary ammonium solution [500 ppm], immersion in hypersaline solution [50 g/L], hydroblasting (physical removal with high- pressure water) and air exposure. The detailing of each methodology tested is described below.

Treatment	Number of tested oysters	Chemical formula	Product concentration used	Exposure time (h)
Freshwater	120	NA	NA	24
Hypersaline water (50 PSU)*	120	NaCl	21 g/L	24
Sodium hypochlorite	120	NaClO	100 mg/L	1
Exposure to air	120	NA	NA	24
Quaternary ammonia	120	CH ₃ (CH ₂)8CH ₂ N + (CH3)2Cl	500 mg/L	1
Hydroblasting	120	NA	NA	NA
Control	120	NA	NA	NA

Table I. Treatments for reducing the colonization of oysters by epibionts. NA= Not applicable; (*) Obtained from the dissolution of refined iodized salt.

For immersion in freshwater (salinity= 0 g/L), ovsters were held for 24 h in 70 L obtained in a local water spring. The treatment in hypersaline water was applied in a tank containing 70 L of the seawater, obtained at the farm area (salinity= 23 g/L). The increase of water salinity (to 50 g/L) was promoted by the addition of iodized salt. The oysters were exposed to the hypersaline solution for 24 h. For the treatment with sodium hypochlorite, a commercial solution (Biotec[®] - 12%) was diluted in freshwater to obtain a final concentration of 100 ppm. The immersion was performed in 70 L of solution for 1 h. A commercial quaternary ammonium solution (Quatermon, Chemitec[®] - 15%) was also diluted in freshwater and the final concentration used was 500 ppm. The immersion was performed for 1 h. In all immersion treatments, the density adopted was 120 g/L. During the freshwater and hypersaline immersion, temperature (TagTemp Stick, Novus[®] Brazil), pH (Sensoglass[®] SP1400, Brazil) and dissolved oxygen (DO) concentration (YSI[®] Pro 20, USA) were monitored every 2 h. During the immersion in sodium hypochlorite and quaternary ammonium solutions, water quality parameters were measured every 10 min.

For the air exposure cleaning methodology, the oysters were kept in crate-type plastic boxes, protected from direct sunlight, for 24 h. The air temperature was recorded every 10 min (data logger, TagTemp Stick, Novus[®], Brazil). Hydroblasting was applied 143 using a 10-mPa high-pressure washer machine (Gong, Italy) directly over the oyster shells until the epibionts were completely removed. The total removal of epibionts was achieved in, approximately, 5 min.

The oysters from the control group were not submitted to any cleaning method. After grouping, the control oysters were kept in the same condition of the tested treatments. The tested treatments did not cause mortality during and short-after exposure (20 min). Oysters were considered dead if they presented open valves and do not display any response to a light knock in their shell.

Post-treatment period

In the post-treatment procedure, oysters from each treatment were held in Japanese lanterns (in triplicate) for 15 and 30 days. To reduce stress by manipulation and air exposure, oysters were handled the minimum necessary during selection and transference to the experimental units (20 min).

As the position on the water column can influence epibiont colonization (Lacoste et al. 2014), oysters were equally distributed on each of the four floors of the Japanese lantern (Figure 1). To facilitate the identification of groups and ensure that the post-treatment conditions were the same for all individuals, the animals were grouped in plastic nets (0.5 cm opening mesh) and identified with numbered seals. The number of oysters used in each group is described in Table II.

Water salinity and temperature were measured daily, through the experimental period (max 30 days). Additionally, the distance of the Japanese lanterns in relation to the bottom at their installation site was estimated. In this case, since there was no specific tide-table for the site of the experiment, the tide table provided by the National Oceanographic Database of the Brazilian Navy (BNDO 2016) for the region of Galheta Canal, Paranaguá Bay, Paraná, Brazil, was used. At the beginning of the experiment, the actual depth of the site was measured at the time the lanterns were installed. The minimum local depth (X) was calculated based on the initially measured local depth (P); the tide height in Galheta Canal at the beginning of the experiment (m1); and the minimum daily tide height (m2) in that place, using the following equation:

$$x = \frac{P \times m1}{m2}$$



Figure 1. General layout of treatments in the Japanese lanterns during the experiment. T = Treatment; C = Control.

Treatment	Number of Japanese lanterns	Number of floors on the Japanese lanterns	Post- experimental period (days)	Number of tested oysters per floor	Number of tested oysters per treatment
Freebwater	2	4	15	5	120
Fleshwater	3	4	30	5	120
the second line waters	2	,	15	5	120
Hypersaline water	3	4	30	5	120
Sodium hypochlorite	2	,	15	5	120
	3	4	30	5	120
	2	,	15	5	10.0
Quaternary ammonia	3	4	30	5	120
	2	,	15	5	10.0
Hydroblasting	3	4	30	5	120
Exposure to air	2	,	15	5	10.0
	3	4	30	5	120
То	840				

Table II. Experimental design adopted to the evaluation of different methodologies for the reduction of epibionts on the mangrove oyster (*Crassostrea gasar*) during cultivation conditions.

Epibionts identification and mortality measurement

At the end of 15 and 30 days, oysters from each treatment (n= 5 /period/treatment) were sampled. At the same time, mortality was evaluated following the same procedure described above. Then, oysters were packed in individual plastic bags, storage in refrigerated thermal boxes, and transported to the Aquatic Organisms Research Laboratory (LAPOA), located at the Federal University of Paraná, in Curitiba/ PR, South Brazil. Epibionts were removed from the shell with the aid of a soft bristle brush under freshwater. The washed material was sieved in 90 µm mesh, fixed and preserved in 4% formalin. Live epibionts attached to the shells, such as barnacles, were quantified immediately after the removing process. All epibionts from each sample were identified and taxonomic quantified under a stereomicroscope (Leica® MZ6, Germany).

Statistical analyses

The number of epibionts in each treatment was log transformed. The role of all categorical variables evaluated (lantern, walking, time, and treatment) on the number of epibionts (log transformed data) were analyzed. Then, the homoscedasticity tests were performed using Cocharn, Hartley and Bartlett and Levene methods, followed by Duncan *post-hoc* analysis. To assess the degree of significance of oyster mortality rates in the different treatments and in the different periods analyzed (15 and 30 days), proportion tests were performed using the McNemar *Chi*-square test for the significance of changes. All analyses were conducted using Statistica[®] 10.0 software (StatSoft, USA).

RESULTS

During the application of the cleaning methods, the average air temperature was 24.6 °C, water temperature varied between 23.6 and 29.6 °C, pH varied between 6.4 and 7.8, and DO were maintained above 4.7 mg/L (Table III). In the postexposure period, the mean water temperature was 27.2 °C on the 1st floor and 26.8 °C on the 4th floor of the Japanese lantern. The average salinity was 23 g/L, ranging from 19 to 30 g/L (Figure 2). The Japanese lanterns remained at an average depth of 2.2 m (1st floor) and 1.6 m (4th floor) from the bottom of the canal during the experimentation period.

Throughout the experiment, a total of 60949 epibionts, belonging to nine phyla, were identified. The most abundant phyla were Arthropoda (Subphylum Crustacea), with 38619 animals (63.4%), followed by Mollusca, with 20246 organisms (33.2%) and Annelida, with 1792 individuals (2.9%). We identified the presence of algae of the Rhodophyta phylum (except for treatments with freshwater immersion, hypersaline water, exposure to air and hydroblasting) and Ectoprocta phylum (all treatments). Their quantification, however, was not possible since only fragments were found.

There was greater prevalence of epibionts in the control group (n= 10180), followed by the

quaternary ammonia-based (n= 9107), sodium hypochlorite (n= 8291), air exposure (n= 7596) and hypersaline water treatments (n= 7367). In addition, the treatments with the fewest epibionts were hydroblasting (n= 4239) and freshwater (n= 5085). The number of epibionts on the oyster shells in the two periods (15 and 30 days) was almost always lower than at Time 0 (n= 4542 organisms), except for the control group [both in 15 (n= 5440) and 30 days (n= 4740)] and for the treatment based on quaternary ammonia (30 days) (n= 4649) (Table IV).

There were significant differences (Duncan test, p<0.001) among the cleaning treatments. Hydroblasting and exposure of the animals to freshwater reduced the number of epibionts when compared to the control. There was no difference in the number of epibionts recorded between the two sampling periods [15 (Duncan test, p = 0.41) and 30 days (Duncan test, p = 0.42)] (Figure 3). Both methods (hydroblasting and freshwater) were also the most effective for reducing organisms belonging to the Annelida, Mollusca, and Arthropoda phyla. Conversely, exposure to air did not demonstrate efficacy for

reatments were not significant (p>0.05). T ₁ : water temperature; T ₂ : air temperature; OD: dissolved oxygen; S: salinity; NA: Not applicable.								
Treatment	т (°С)	т (°с)	nH	OD(mg/I)	S (PSII)			

Table III. Abjectic variables measured during the exster cleaning phase. The ebsenved differences between the

Treatment	Т ₁ (°С)	T ₂ (°C)	рН	OD (mg/L)	S (PSU)
Freshwater	24.1 (23.6-25.2)	-	7.0 (6.4-6.6)	5.9 (4.7-6.2)	0
Hypersaline water	26.3 (24.7-29.6)	-	6.9 (6.7-7.8)	6.5 (6.3-6.7)	50
Sodium hypochlorite	26.3 (26.1-26.5)	-	6.8 (6.8-6.9)	6.2 (6.1-6.3)	0
Quaternary ammonia	26.4 (26.3-26.6)	-	6.7 (6.7-6.8)	6.5 (6.4-6.5)	0
Hydroblasting	-	-	NA	NA	0
Exposure to air	-	24.6 (23.4-24.9)	NA	NA	NA

any of the phyla found when compared to the control group.

None of the six treatments were effective in reducing the number of Platyhelminthes, Rhodophyta and Ectoprocta recorded on the oyster shells. In three of the nine phyla identified it was not possible to test the effectiveness of the treatments in relation to the control, due to the low occurrences (Chordata: n= 2; Cnidaria: n= 36; and Echinodermata: n= 1) (Table V). The position of the oyster on the water column did not cause differences on the total number of epibionts (Duncan test, p= 0.68).

At the end of the experiment (30 days), the mortality rate of treated oysters was 8.8%, and in the control group was 22.5%. There was a significant increase (McNemar *Chi*-square p= 0.003) in the number of dead oysters between 15 and 30 days in the control compared to the treatments with air exposure and hydroblasting (Figure 4).

DISCUSSION

During both exposure and post-exposure periods, variations in water guality parameters were observed. Nevertheless, all measured values were within the ranges considered optimal or tolerable by oysters as follows: water temperature (23-31 °C) (Ansa & Bashir 2007), air temperature (5-25 °C) (Zhang et al. 2006), pH (6.7-8.7) (Morales 1986), dissolved oxygen (>3 mg/L) (Mello 2007), and salinity (10-50) (Funo et al. 2015). Although we identified the presence of epibionts from nine phyla, a higher occurrence of Arthropoda, Mollusca, and Annelida was detected. The result agrees with the results obtained by Schuster-Pinto (2007) in Guaratuba Bay. In this work, the author identified higher prevalence of epibionts from Mollusca and Arthropoda phyla on commercialized Crassostrea sp. The high prevalence of animals from the Arthropoda phyla is due to the presence of cirripeds (barnacles). Barnacles attach themselves to solid surfaces (e.g., shells



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and cultivation structures) and live in dense agglomerations in the same places occupied by oysters, competing with them for food and oxygen (Ruwa & Polk 1994). According to Alvarenga & Nalesso (2006), barnacles can affect oyster filtration and consequently their survival rates. In addition, barnacles can damage the growth border and make it difficult for oysters to open and close their valves. The high abundance of Mollusca is due to the mytilids, mainly members of the genera Mytella and Sphenia. According to Galvão et al. (2009), when fixed on oyster shells or cultivation structures these animals secrete the byssus, a substance of high viscosity which is able to cause local sludge accumulation and can lead to the death of oysters. In contrast, a study conducted by Pereira et al. (1988), in a 271 location relatively close to the Bay of Guaratuba, they found that even high densities of mytilids encrusted on oysters did not cause mortality or reduction in the growth of oysters cultivated on a table system. The third most dominant phylum

of epibiont found on the oysters was Annelida, which was represented by a high number of polychaetes, mainly errant and tubicolous forms. According to Schuster-Pinto (2007) polychaetes are often found in abundance on bivalves in the area of the present study. Additionally, Figueras & Villalba (1988) reported that some polychaetes are considered parasites of oysters and can cause mortality, especially in younger individuals.

The oysters from the control group presented the highest number of epibionts on their shells. This result was not unexpected as the control group did not undergo any type of cleaning procedure during the three months preceding or during the experimental period. In addition, control oysters, as well as those exposed to air, showed increasing mortality rates between the 15th and 30th days of the experiment. These data corroborate with the findings of Jakob & Wang (1994), who reported a higher mortality rate of oysters (*Crassostrea*

		Treatment													
Phylum		Сог	ntrol	Fresh	water	Soc hypoc	lium hlorite	Hype wa	rsaline Iter	Expo to	osure air	Quate amm	ernary Ionia	Hydro	blasting
Time	0	15	30	15	30	15	30	15	30	15	30	15	30	15	30
Arthropoda	3348	3103	2896	1418	1813	2158	2794	2353	2834	2312	2344	2730	2,746	1011	1411
Mollusca	1146	1641	1890	807	917	1347	1742	816	1174	1149	1417	1644	1754	800	856
Annelida	30	177	429	31	73	44	174	20	130	117	212	50	129	38	108
Chordata	0	0	1	0	0	0	0	0	1	0	0	0	0	0	0
Platyhelminthes	18	19	21	7	14	12	13	14	19	22	16	34	16	0	10
Cnidaria	0	0	3	0	5	0	7	0	6	0	7	0	4	1	3
Echinodermata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Rhodophyta	А	Р	Р	A	Р	Р	Р	А	Р	Р	А	Р	Р	А	А
Ectoprocta	Р	Р	Р	Р	Р	Р	Р	Р	Р	Р	Р	Р	Р	Р	Р
Total	4542	5440	4740	2263	2822	4085	4206	3517	3850	3600	3996	4458	4649	1850	2389

 Table IV. Number of epibionts on the oyster shells submitted to different cleaning treatments after 0, 15 and 30 days.

METHODS REDUCING EPIBIONTS

aiaas) when they were not submitted to a regular cleaning process. On the other hand, Schuster-Pinto (2007) found no relationship between mortality rates and frequency of shell cleaning. According to Sala & Lucchetti (2008), oyster farmers usually report difficulties and limitations during cleaning treatments, as these procedures require the removal of the cultivation structures from the water and their transportation to the management sites before cleaning occurs. In fact, cleaning procedures require high physical effort and are timeconsuming, causing farmers to extend the period between its application which, usually, results in a reduction of growth rates and consequently economic losses (Manzoni 2001). Thus, the use of easily and readily applied techniques, as those described in the present work, could facilitate adhesion by farmers as they are also safe for the operator, for the oysters and for the environment. Our results, however, evidenced that all the methods employed in the cleaning of oyster shells presented some inefficiency, both in relation to the numerical reduction of the epibionts and in relation to the identified

zoological groups, since none of them allowed the complete elimination of the organisms during the period of exposure. The air exposure, for example, is a method commonly used by farmers to reduce epibionts from oyster shells (Adams et al. 2011), but in the present study, it proved to be inefficient regarding reducing epibionts and avoiding oyster mortality. Gervis & Sims (1992) reported that air-exposed oysters may show different responses to treatments because of the composition of the epibiont community on their shells (rigid bodies or soft bodies) and the sensitivity of each species of oyster. According to Fitridge et al. (2012), some limestone organisms (such as barnacles) can maintain their internal moisture for long periods and consequently do not die during exposure to air. Thus, is reasonable to assume that lack of efficiency presented by the air exposure method is related to the prevalence of barnacles attached to the oyster's shells.

Chemical treatments (exposure to hypersaline water, sodium hypochlorite, and quaternary ammonia) would probably have provided better results, at least in terms of



elimination of epibionts, if the exposure time or the concentrations used had been higher than those used. However, this would run counter to our purpose of testing chemical methods that could act faster and be more efficient than conventional options. In addition, the application of chemical methods is financial higher and infer environmental risks, which are directly associated with the quantities of products used and the effluents generated. According to Williams & Schroeder (2004), the use of sodium hypochlorite for reduction of epibionts should involve a protocol that is fast and rigorously controlled, as this product is potentially toxic to the general animal communities and to the environment. Rolheiser et al. (2012) observed that the exposure to salinities of 40, 50 and 70 g/L did not cause a significant decrease in the number of epibionts on the oyster shells. Additionally, Denny (2008) found that exposure to sodium hypochlorite at a concentration of 0.5% for two minutes reduced the propagation of the ascidians *Didemnum vexillum*, but did not reduce the presence of tunicates on the shells of the mussel *Perna canaliculus*.

The immersion of oysters in freshwater showed relatively high efficiency in the reduction of epibionts, a result which differed from that reported by Rolheiser et al. (2012), who evaluated methods for reducing biofouling in C. gigas. In that case, the authors recorded a decrease in the survival of oysters exposed for only ten minutes to freshwater and concluded that there was no reduction in the number of epibionts. In the present study, as C. gasar exhibited a high survival capacity of approximately 15 days in freshwater (Horodesky et al. 2018), this has proved to be a safe and non-residual method for epibiont control. Hydroblasting and freshwater immersion were the most efficient methods of reducing the number of organisms in the Mollusca and Arthropoda phyla. This finding

Table V. Analysis of effectiveness in the removal of different epibionts found in oyster shells compared to the control group. Yes = the existence of significant differences; No = no significant differences; NC = not comparable; α = 0.05.

	Treatment										
Phylum	Hydroblasting	Fresh water	Treatment Quate Hypersaline water Sodium hypochlorite Quate Yes Yes Ye (<0.00) (<0.00) (<0.00) Yes No No Yes No No Yes No No (<0.00) (1.0) (1.0) (<0.00) (1.0) (1.0) No No No No No No (0.26) (0.62) (0.7 No No No No No No No No No (0.25) (0.38) (0.4 (0.39) (0.51) (0.51) NC NC No NC NC No	Quaternary ammonia	Exposure to air						
Annelida	Yes	Yes	Yes	Yes	Yes	No					
	(<0.00)	(<0.00)	(<0.00)	(<0.00)	(<0.00)	(1.0)					
Mollusca	Yes	Yes	Yes	No	No	No					
	(<0.00)	(<0.00)	(<0.00)	(1.0)	(1.0)	(0.18)					
Arthropoda	Yes	Yes	No	No	No	No					
	(<0.00)	(<0.00)	(1.0)	(1.0)	(1.0)	(0.54)					
Platyhelminthes	No	No	No	No	No	No					
	(1.01)	(0.38)	(0.26)	(0.62)	(0.77)	(0.26)					
Rhodophyta	No	No	No	No	No	No					
	(0.27)	(0.54)	(0.55)	(0.38)	(0.41)	(0.33)					
Ectoprocta	No	No	No	No	No	No					
	(0.52)	(0.31)	(0.39)	(0.51)	(0.32)	(0.50)					
Chordata	NC	NC	NC	NC	NC	NC					
Cnidaria	NC	NC	NC	NC	NC	NC					
Echinodermata	NC	NC	NC	NC	NC	NC					



Figure 4. Cumulative percentage of dead ovsters in the control group and in the respective treatments during the experiment for epibiont control. **Capital letters indicate** differences (Duncan test; p<0.05) among treatments and lowercase letters indicate differences between 15 and 30 days (McNemar Chisquare test; p<0.05).

corroborates the study of Mcennulty et al. (2001), who cite both methods as being the most widely used in the reduction of small mollusks and arthropods (mainly crabs) on oyster shells. Santana (2005) stated that hydroblasting is a method widely used by oyster farmers along the Santa Catarina coast, the main oyster producing area in Brazil. Hydroblasting is used both for the cleaning of the animals, as well as for the culture structures. According to this author, this cleaning method increases food flow through the protective screens, resulting in higher oyster growth and survival rates during a production cycle. Forrest and Blakemore (2006) also indicated that hydroblasting is a high-efficiency non-selective method for oyster cleaning.

The exposure of mangrove oysters (*C. gasar*) to freshwater for 24 h and the cleaning of their shells by hydroblasting were the most effective methods for controlling the number of epibionts. These methods also presented minimal mortality losses associated with management process during oyster farming.

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The study was designed by all AO and AH and GGCW. The experiments and measurements were performed by AH, GDP and NC. AO and AH wrote the first draft of the manuscript, GDP, NC and GGCW edited it, and all authors approved the final version.

