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Breeding and Larval Rearing in Hatchery of *Macrobrachium Vollenhovenii* (Herklots, 1857) In a Perspective of Biological Control against Human Schistosomiasis in Tropical Area

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Abstract

As part of an effort to use biological control against human schistosomiasis, post-larval production of the prawn *Macrobrachium vollenhovenii* was carried out in a hatchery. This species is a freshwater native and is fond of mollusks, intermediate hosts of schistosomes that transmit human diseases in the wild. The larval production of this species occurred in six (06) tanks - two 200 L and four 600 L. The management of brood stock took place in fresh water, with a ratio of one male to four females. Each fertilized female produced thousands of eggs attached to the abdomen until hatching about two weeks later. The larvae resulting from this reproduction were raised to the post-larval stage in brackish water at a salinity of 15 ppt. The larvae must undergo a molt between each stage. We identified 15 morphological stages between stage 1 and the post-larval stage. The duration from stage 1 until onset of the first post-larval stage in the six (06) tanks was on average 54 days after hatching. The success of larvi culture is defined by the rate of metamorphosis of the larvae into post-larvae, which, in our case, varied between 0.17% and 3.08%, depending on the production tanks, with an average of 2.8%. These results show that the production of *M. vollenhovenii* in hatchery is possible, presenting a possibility for aquaculture production of this species as well as for biological control against schistosomiasis in a tropical area.

Keywords: Reproduction; *Macrobrachium vollenhovenii*; Larvication; Post-larvae; Human schistosomiasis

Introduction

Macrobrachium prawns are used for aquaculture purposes in many countries around the world. Species of this genus, including *Macrobrachium vollenhovenii*, act as agents against human schistosomiasis by feeding on a variety of mollusks that are intermediate hosts of schistosomes responsible for schistosomiasis in humans (Sokolow *et al.,* 2013; Swartz *et al.,* 2015; Savaya *et al.,* 2017)[1-3]. *Macrobrachium vollenhovenii* is a freshwater prawn endemic to the West African coast, from Senegal to Congo (Zabi et Le Loeuff, 1992)[4]. It is an exoskeletal crustacean that frequently needs to molt to grow and reproduce. Because its carapace is very rich in calcium, it consumes the shells of mollusks, intermediate hosts of trematodes responsible for human schistosomiasis, to compensate for calcium losses after each molt. Hence, it helps to stop the cycle of disease transmission by eliminating mollusks as intermediate hosts of schistosomes (Sokolow *et al.,* 2013)[5]. *M. vollenhovenii* is part of the fauna of benthic and littoral zones of the intertropical zone of West Africa from northern Senegal to southern Congo (Zabi et Le Loeuff, 1992)[4]. It is an indigenous species that was historically abundant in the Senegal River. It is today endangered in this ecosystem, particularly in the upstream part of the Diama dam. Its scarcity began in the 1990s, fol-

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lowing the installation of the Diama dam on the Senegal River in 1986 (Southgate, 1997)[6]. This dam was created to facilitate land management and the development of irrigated agriculture year-round in the upstream part of this dam.

The implementation of the Diama dam limits the salt-water intrusion toward the upstream part of this dam (www.omvs.org) and has resulted in the upset of the Senegal River ecosystem through the modification of ecological conditions. These environmental changes restricted the migratory movements of amphibiotic species on both sides of Diama dam (DeGeorges and Reilly, 2006)[7]. *M. vollenhovenii* is a catadromous species that lives in freshwater and breeds in an estuarine environment in around 15 ppt salty water. The change in the environment of the Senegal River threatens the survival of this species, which can no longer effectively function as a predator and regulator of mollusk populations. As a result, the prevalence and intensity of schistosomiasis increased rapidly in the villages and surrounding towns of the Senegal River after the completion of the Diama dam (Southgate, 1997)[6].

The purpose of this work is the production and rearing of the larvae until post-larvae stage of *M. vollenhovenii* prawns in the hatchery in order to be able to repopulate the natural environment with this species and to contribute to the biological control against schistosomiasis.

The few studies carried out on this species have focused mainly on its biology and ecology (Savaya *et al.,* 2014; Konan *et al.,* 2010; Bello-Olusoji, 1997; Anetekhai, 1986; Marioghae, 1982)[8-12]. Specific breeding techniques for *M. vollenhovenii* are poorly studied around the world (Niass *et al.,* 2015; Makombu *et al.,* 2014; Willführ-Nast *et al.,* 1993; Anetekhai *et al.,* 1987) [13-16]. This study aims to control the production techniques and reproduction of post-larvae of *M. vollenhovenii* in the hatchery.

Material and Methods

Fishing on the Senegal River and transport of *M. vollenhovenii*' broodstock

The *M. vollenhovenii* individuals were fished in the Senegal River within a radius of one km on both sides of the Diama dam. The coordinates of some fishing spots were taken using a GPS (GPS eTrex 10, GARMIN) and visualized in Google Earth (Figure 1). The Diama dam is located about 42 km from Saint-Louis. The broodstock caught with a 14 mm mesh net were stored on site in a cage submerged in the river. When they were recovered, they were put in basins previously filled with water from the river and transported to the shore. To ensure safe conditioning for transport, the bags or carrying bag were filled with one-third water from the hatchery where the parents were stored, and one-third water from the river. Depending on the volume of the bags and the size of the animal, four (04) or five (05) *M. vollenhovenii* broodstock were placed in each bag.

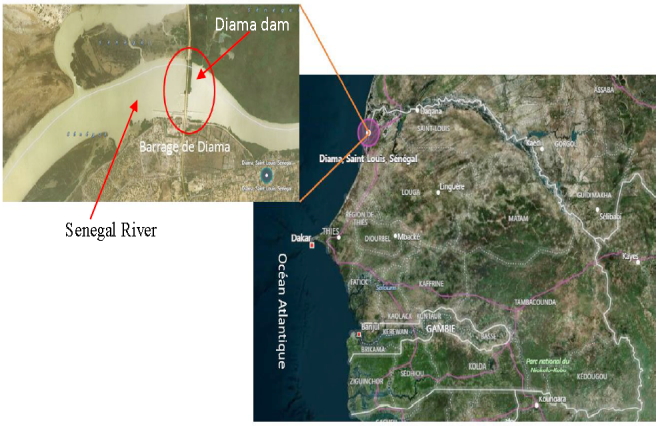

Figure 1: Geographical location of *M. vollenhovenii* fishing area on the Senegal River (Google Earth image)

For the oxygenation of the transport water, a bottle of oxygen was used by placing a fitting on the bag. The air inside the bag was first completely expelled. Then, the oxygen cylinder was moderately opened to put a reserve of oxygen.

Once filled, the bag was sealed to prevent oxygen from escaping. The pouches were double-bagged to prevent broodstock from using their long and pointed chelipeds from piercing them and causing the loss of the water and oxygen inside. The securely closed bags were laid horizontally in polyester cases and transported to the hatchery.

Hatchery

The hatchery of the "Aquaculture Pour La Santé" (APLS) project was located in the city of Saint-Louis (Senegal) with an area of approximately 200 m^2. It is there that the breeding, reproduction and production of *M. vollenhovenii* post-larvae took place between May 2015 and December 2016. The infrastructure consisted of two main units: the *M. vollenhovenii*' larval production unit and the unit of live prey culture.

Larvae production unit: This consisted of 118 m^2 of the hatchery and included all the breeding tanks for the broodstock and larviculture of *M. vollenhovenii*. The broodstock rearing tank was 8m^3. The larvication tanks are of different shape, color and volume. They include seven (07) 200 L blue tanks, numbered from B1 to B7 and each connected to a biological filter of 45 L; five (05) 600 L white tanks numbered from C3 to C7 with each connected to a 100 L biological filter. The tanks numbered C0 to C2 had a capacity of 1 m^3 each and had different uses. The tank C0 was used as a reservoir of sea water and the tank C1 was used for storing water and degassing chlorine from tap water. The tank C2 was used for the maturation of berried females from the broodstock.

Live prey unit: The live prey culture unit was located in 5 m^2 room. It hosted the conical-bottomed incubation tank of the *Artemia* used in feeding larvae. Each conical-bottomed tank had a 150 W immersion heater to maintain the incubation temperature of the *Artemia* at 28°C. Also, an air stone was immersed in each conical-bottomed tank to improve the oxygenation in the culture area. The room was permanently lit with five (05) neon lamps of 12 V each for the light needs of the *Artemia*.

Rearing broodstock

The broodstock were raised in a tank of 8 m3, half filled with tap water previously dechlorinated. The sex ratio was one (01) male per four (04) females. 70 broodstock were put in the breeding tank including 14 males with an average weight of 40 ± 4 g and 56 females with an average weight of 28 ± 3 g. Five (05) immersion heaters of 300 W each set at a maximum temperature of 32°C, were connected to the tank to keep the water temperature between 28 and 30°C to induce sexual maturation of the gonads in females. The tank was covered with a tarpaulin to prevent heat loss caused by exchanges with the exterior environment. The water temperature was checked daily using an oxymeter (YSI Pro 2030). The rise of water temperature in the tank induces the maturation of the gonads in the females, which become bright orange and very visible through the shell (Figure 2).

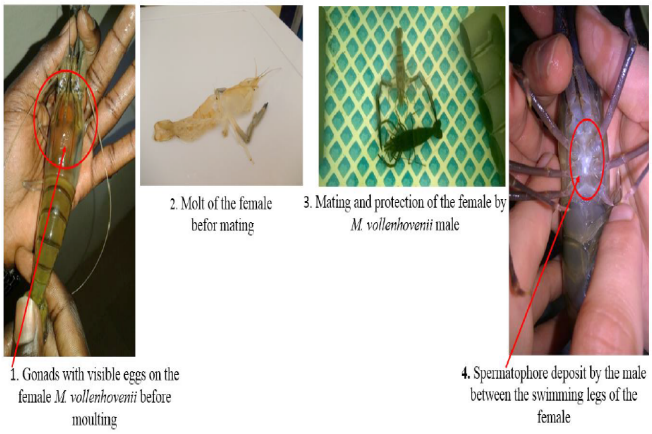

Figure 2: Mating process of *M. vollenhovenii*

Feeding broodstock

Three types of food were combined to feed the broodstock: i) a food formulated locally by the National Aquaculture Agency (ANA) from local by-products (peanut cake, rice bran, fish meal, fish oil, cuttlefish shell, etc.); (ii) an industrial feed imported from the United States of America (USA); and (iii) local shrimp purchased on the local market to supplement the calcium content. This combination of foods was justified by the high cost of the imported food and the need to reduce production costs. The spawners were fed once by day at 5% of their biomass in pellet feed and shrimp purchased at the local market and cut into small pieces.

Molting and mating of broodstocks

Crustaceans are organisms with exoskeletons, an external skeletal framework. This frame allows them to protect themselves against predators but, in some cases, prevents them to grow or even to mate. The *M. vollenhovenii* is a closed thelycum species, which means that females of this species required to get rid of their exoskeleton by molting in order to mate with a male (Figure 2).

The female heeds her shell by bending ventrally, pulling her abdomen out of the carapace first and her cephalothorax last. After molting, one of the males comes close to the female and, using its pereiopods, hangs underneath the abdomen to place his sexual appendage on the female's thistle. The male deposits his sperm as a gelatinous mass on the ventral thoracic region between the pereiopods (walking legs) of the female and stays close to it for some time to protect it from cannibalism. During this period, she is vulnerable to predators due to the loss of her carapace (Figure 2).

After mating, females with eggs emerged from the breeding tank and were individually placed in cages in another tank to monitor the evolution of egg color and the different stages of embryogenesis.

Hatching of larvae

Macrobrachium vollenhovenii larvae live in saline brackish water of about 15 ppt (Willführ-Nast *et al.,* 1993)[15]. The water from the larvication tanks was maintained at the same salinity value. The females were acclimatized before the hatching of the larvae by progressively transitioning basins from freshwater to brackish water. The salinity was gradually increased by 1 ppt per hour up to 7 ppt. After hatching, the salinity of the water containing the larvae was gradually increased again by 1 ppt per hour until reaching 15 ppt.

After acclimatization, the larvae were transferred from the hatching tanks to the larvication tanks where the larval rearing took place.

Breeding larvae of *M. vollenhovenii*

After transferring the larvae to tank groups B (B4 and B5) and C (C4 to C7), the number of larvae obtained by hatching was estimated as larval density (LD) per liter as (Makombu *et al.,* 2014)[10]:

$$LD=n/Nx2$$

With n = number of larvae counted in a 500 ml sample; N = number of 500 ml samples where the number of larvae were counted.

Every ten days the density of each tank was estimated again in order to follow the variation of larval density.

The identification of larval stages of *M. vollenhovenii* was made using a compound microscope ((Leica BIOMED magnification x10) based on the work of Liu and Liao (1999), who identified 15 larval stages for this species.

To track larval growth in each tank, the Index of Stage Larval (ISL) was estimated. A sample of six (06) larvae was taken in each tank every five days and observed under a microscope. The stage of development of each larva was noted and the ISL was calculated as follows according to Makombu et al. (2014)[14]:

$$ISL=\Sigma SiniSi/N$$

With Si = larval stage; ni = number of larvae observed by larval stage; N = Total number of larvae observed.
Plotting the variation of the ISL allowed us to determine the date of appearance of the first post-larval (PL) stages.

Larval diet

Also, three types of food were combined for feeding larvae: powdered artificial food imported from the USA, fresh shrimp and *Artemia*. The *Artemia* were the main source of food used for larvae. The boxes of *Artemia* were imported from the USA

(strain: INVE *Artemia* cysts). The *Artemia* were in the form of cysts packaged in airtight boxes and stored at a temperature of 5°C. To use them as food, the *Artemia* were prepared in incubation cones where they hatch in a 35 ppt salt water solution under an incubation temperature of 28°C (Figure 3). The cones were well oxygenated and illuminated to ensure a good hatching rate; i.e., more than 80% of the *Artemia* nauplii. After 21 hours of incubation, the *Artemia* nauplii were carefully harvested and separated from empty shells before giving them to the larvae. To allow the larvae to maximize nutrition by spending less energy searching for prey, a density of 5 *Artemia* nauplii per 1 ml of water in the tanks was maintained.

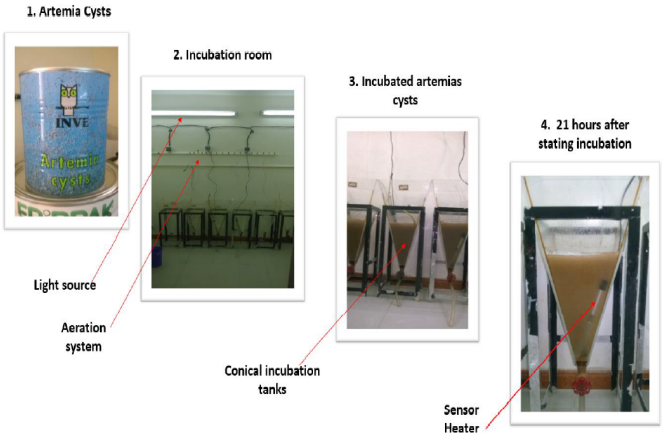

Figure 3: Preparation of *Artemia* for feeding *M. vollenhovenii* larvae

Larval water parameters monitoring

In order to maintain the good water quality in the breeding tanks, the physio-chemical parameters were regularly monitored from the beginning of the larvication period until the metamorphosis of the larvae into post-larvae stage.

Parameters such as temperature, salinity, pH, NH_4, NO_2 and NO_3 were taken twice a week using an electro-photometer kit (YSI 9300).

Tank water monitoring

The larval culture tanks were siphoned daily in the morning before feeding the larvae. After siphoning, live larvae siphoned into the tanks were harvested and returned to the production tanks. The strainer that is on the overflow of each tank was also disassembled, cleaned and disinfected in a chlorinated water solution before the water renewal in each tank. The decision to perform water renewal of the tanks was made according to the physio-chemical parameters of the water passing through the values, measured using the electro-photometer. If the ammonium test showed that the level was up to 0.09 $mg.L^{-1}$, according to the indicative value given by Zweig et al. (1999)[17], half of the water in the tank was renewed.

Statistical analyses

Data on physio-chemical parameters were compared between tanks by the one-way ANOVA test using statistical software R (version i386 3.4.0). All statistical tests were concluded at the significance level $\alpha = 0.05$.

Results

Water quality

The means and standard deviations of the physiochemical parameters obtained from monitoring the water quality of the larval culture tanks are given in Table 1. The average water temperature of the six tanks was between 27.6 ± 0.23 and 28.1 ± 0.18°C. The pH varied between 7.5 ± 0.01 and 7.9 ± 0.03 and the salinity was almost stable at 15 ppt in all tanks. The statistical tests showed no significant variation in temperature and pH values between the six tanks (p-value > 0.05). However, ammonium and nitrite showed significant variations between different tanks. The ammonium concentrations were 0.07 ± 0.04 $mg.L^{-1}$ for tank B4 and 0.3 ± 0.15 $mg.L^{-1}$ for tank C5. For nitrite, the highest concentration was noted in tank C5 (0.4 ± 0.24 $mg.L^{-1}$) and the lowest in tank B5 (0.2 ± 0.11 $mg.L^{-1}$). For the other tanks, the nitrite concentration remained below 0.3 $mg.L^{-1}$. The ammonium and nitrite concentrations were found to be significantly different between larval culture tanks (*p*-value < 0.05).

Fertilization and maturation of *M. vollenhovenii* eggs

Females fertilized by males began carrying eggs under their tails 8 hours after fertilization. The female deposits her eggs underneath her abdomen after molting and mating. Egg color changed from orange to dark brown, corresponding to different embryonic stages, before larval hatching (Figure 4). Depending on the maturation of the eggs, cell multiplication resulted in the formation of some organs that are visible in the eggs. The appearance of the eyes in the form of small black spots was observed under a microscope beginning on the 5th day after fertilization (Figure 4). The eyes become more visible on the 12th day. At this step in development, the tail begins to curve towards the head in the embryonic sac. On the 50th day after fertilization, the microscopic observations showed the heartbeat in the larvae. The hatching of the larvae took place around the 60th day after fertilization.

Table 1: Averages and standard deviations of physiochemical parameters of larvication tanks

Parameter	Bac B4	Bac B5	Bac C4	Bac C5	Bac C6	Bac C7
Temperature (°C)	27.8±0.17	28.1±0.18	28±0.09	27.9±0.1	27.6±0.23	28.1±0.13
pH	7.8±0.015	7.7±0.016	7.5±0.012	7.9±0.017	7.6±0.013	7.6±0.013
Salinity (ppt)	15.01±0.03	15.05±0.09	15.10±0.16	15.08±0.77	15.08±0.77	15.07±0.12
Ammonium ($mg.L^{-1}$)	0.07±0.04	0.2±0.01	0.08±0.04	0.3±0.15	0.09±0.05	0.2±0.07
Nitrite ($mg.L^{-1}$)	0.2±0.17	0.2±0.11	0.3±0.3	0.4±0.24	0.2±0.14	0.4±0.02

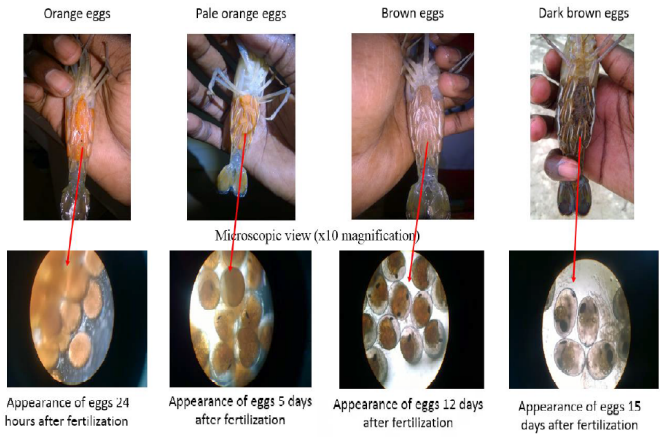

Figure 4: Maturation of eggs and corresponding embryogenesis

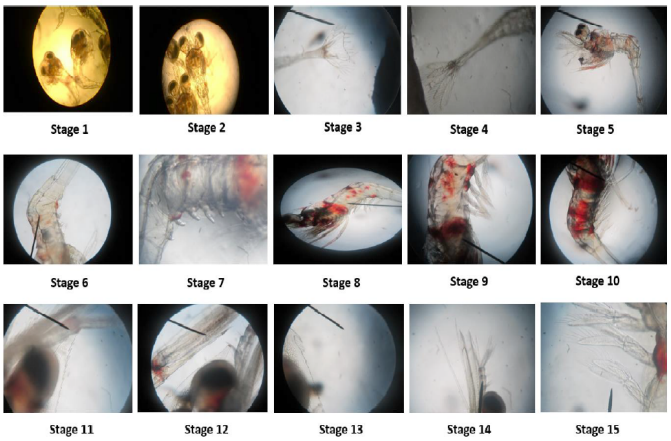

Figure 5: Identification of the different stages of larval development in *M. vollenhovenii* under the microscope (x 10). **Stage 1:** The compound eyes attached on the orbit; **Stage 2:** The compound eyes stalk formed and separated from the orbit; **Stage 3:** the 1st uropod formed and with buds at the base of 2nd uropod; **Stage 4:** 1st and 2nd uropod formed completely: trapezoid telson formed; **Stage5:** the tip of 2nd uropod reached the base part of the telson; **Stage 6:** the rectangular telson formed and the 4th pereiopod opened; **Stage 7:** the 2nd uropod's tip (except setae) reached at the mid part of side spines of telson; **Stage 8:** length of 2nd uropod's setae to be 3 times compared with side spines of telson; the 5th pereiopod much longer than that at the prior stage; **Stage 9:** the trapezoid telson elongated; the buds on the base of 1-5 pleopods occurred; **Stage 10:** the tip of telson formed: 5 pleopods grew out and formed some setae on the forks; **Stage 11:** the left appendage with 5 and right appendage with 3 segments of antennule; 4-6 setae formed on the upper edge of rostrum; with 11-12 segments in the antenna; **Stage 12:** with a lot of branched fine setae on the upper edge of the rostrum; **Stage 13:** the right appendage of antennule with 4 segments; 15 segments in the antenna; **Stage 14:** 6 setae existed between the first and second spine on the upper edge of rostrum; 20 segment in the antenna; **Stage 15:** 6-7 setae occurred on the lower edge of rostrum and 25 segments in the antenna. After the stage 15 the larvae molt and become a post-larvae and get all morphological characters of the adult prawn.

Larval density and cycle of *M. vollenhovenii*

The results on the densities are shown in Table 2. The metamorphosis of the larvae from one stage to other results in mortality that leads to a reduction in the density of larvae produced in all tanks. Almost 50% of the larvae died after 50 days of production. At the end of the experiment, the lowest densities per liter are noted in tanks C4, C7 and B4 and the highest density in tank C6.

The larvae of *M. vollenhovenii* metamorphosed through different stages. A total of 15 larval stages were observed and identified under the microscope. They were photographed and are shown in Figure 5.

Index of Larval Stages (ISL)

The ISL allowed us to follow the growth rate of the larvae in each culture tank.

The larvae monitored in tank B4 showed a very fast and homogeneous growth. The first post-larva stage (PL) was obtained only 31 days after hatching. The ISL showed almost uniform larval growth in this tank. More than 50% of the larvae moved

Table 2: Variation of the larval density estimated on the tanks according to the number of days (D) after hatching (density = number of larvae by liter)

Parameter	Tank B4 (200 L)	Tank B5 (200 L)	Tank C4 (600 L)	Tank C5 (600 L)	Tank C6 (600 L)	Tank C7 (600 L)
D1	53	108	102	70	96	107
D10	49	91	90	66	247	75
D20	48	82	80	62	244	65
D30	44	79	70	60	210	57
D40	39	41	57	32	180	27
D50	28	40	47	24	84	19
D60	23	8	45	16	72	2
D70	4	5	1	12	39	1
D80	1	2	1	2	5	1

to the post-larval stage about 50 days after hatching (Figure 6). The growth of the larvae in tank B5 was not the same as those of tank B4. In this tank, the larvae growth was a little slower. The first post-larval stage was obtained on the 65th day after hatching. The ISL showed stunted larval growth at stage 5 between the 10th and 25th days after hatching (Figure 7).

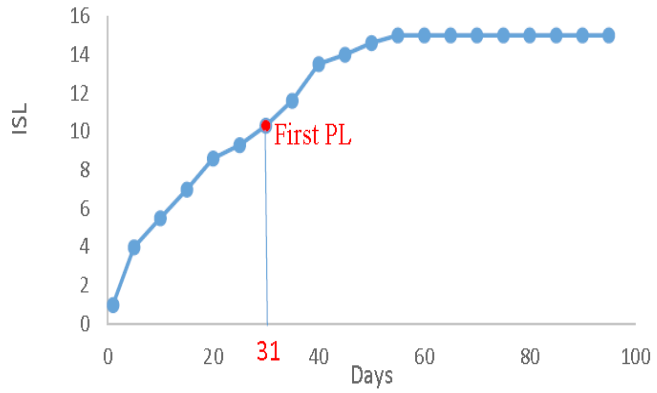

Figure 6: Variation of the index of larval stages (ISL) in terms of time in tank B4 (PL = post-larvae)

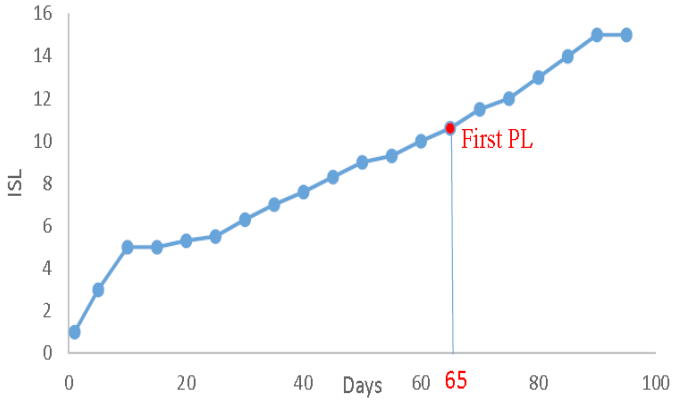

Figure 7: Variation of the index of larval stages in terms of time in tank B5

The larvae growth in tank C4 was very fast and uniform. The variation model of the ISL is almost linear. The passage of time from one larval stage to another did not take more than five days. The first PL stage appeared after 45 days after hatching. The metamorphoses of the larvae into the post-larvae quickly followed the first PL stage. More than 50% of the larvae became PL in less than 10 days after the appearance of the first post-larvae (Figure 8).

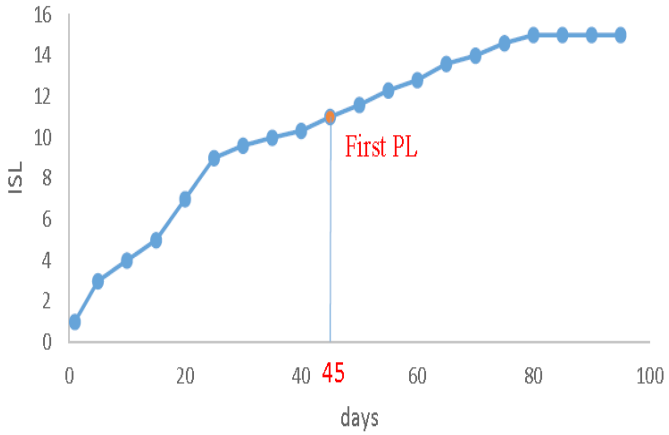

Figure 8: Variation of the index of larval stages in terms of time in tank C4

In tank C5, the larvae show stunting at stage 5 between the 15th and 25th day. Beyond the 25th day, growth resumed very slowly and passed from stage 5 to stage 9 in 35 days of production. The first post-larval stage was obtained on the 60th day and almost all the larvae had metamorphosed into PL after 90 days of production (Figure 9).

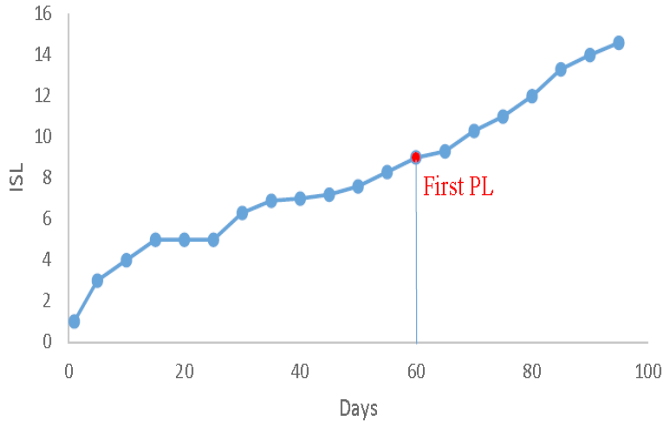

Figure 9: Variation of the index of larval stages in terms of time in tank C5

For tank C6, larval growth was fast and almost uniform. Two slight growth stops were observed. A first was noted at stage 5 between the 15th and 20th day and a second at stage 9 between the 55th and 60th day after hatching. The first PL stage appeared on the 70th day of production. The growth was very rapid beginning on the 10th day, with more than 50% of PL stages being reached 20 days after the appearance of the first PL stage (Figure 10). In tank C7, the larvae had a medium growth rate. A growth stunting was noted at stage 5 in this tank. The transition between stages 8 and 9 was slow. The first PL stage appeared on the 56th day after hatching. Beyond the 56th day, the growth became faster again and all the larvae became PL 20 days later (Figure 11).

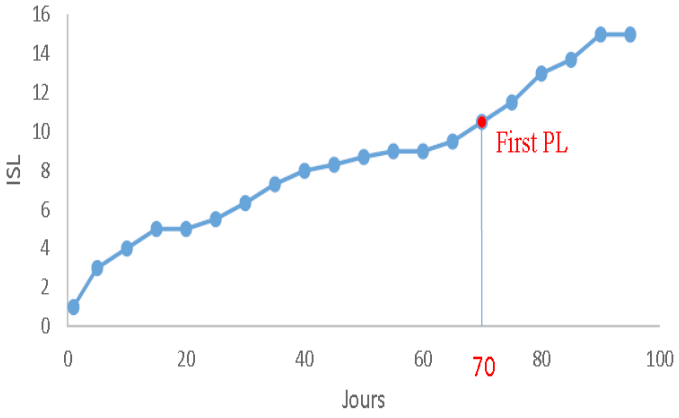

Figure 10: Variation of the index of larval stages in terms of time in tank C6

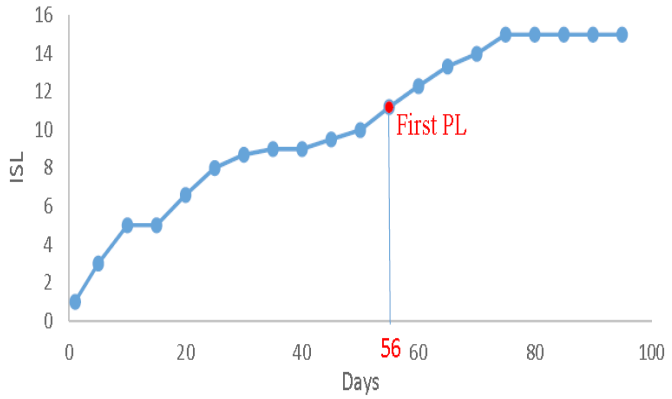

Figure 11: Variation of the index of larval stages in terms of time in tank C

By combining the six larval production tanks, the average duration of onset of the first PL stage was found to be around the 54th day after hatching. The density monitoring provided insight into the survival rate of the larvae within each tank. The rate of post-larvae obtained is greater in tank B4 with 3.08% followed by C5 (2.50%), C6 (2.43%) and C4 (2.18%). The lowest survival rates of larvae metamorphosed to the PL stage were noted in tanks B5 and C7 with 1.50% and 0.17%, respectively.

Discussion

The water temperature for optimal development of the species of the genus *Macrobrachium* was estimated in around 28°C (FAO, 2002). Willführ-Nast et al. (1993) are the first to have tested the breeding of *M. vollenhovenii* in hatchery. According to these authors, the growth of *M. vollenhovenii* 13 larvae was optimal at 28°C. In this study, the water temperatures of the experimental tanks were maintained around this temperature value without major difficulty. However, this high water temperature in the production tanks had negative effects on the quality of the breeding water through changes in chemical parameters such as pH, ammonium, nitrite and nitrate levels. In fact, a high temperature of the rearing area leads to a more intense metabolic activity of the individuals, and consequently to larger waste discharges in water and higher O2 demands (Zweig et al. 1999).

Larvae of *Macrobrachium* genus are highly sensitive to changes in pH, ammonium, and nitrite. To ensure good production of prawns in a hatchery environment, the pH of the water must be maintained between 7.5 and 8.5 (Zweig et al. 1999)[17]. The ammonium, which is very lethal to larvae, must also remain below 0.09 mg.L-1 and nitrite below 0.5 mg.L-1 (Zweig *et al.,* 1999)[17]. In some of our production tanks, the average concentrations of ammonium and nitrite were above these values. The ammonium levels in tanks C5, C7 and B5 were 0.3 ± 0.15 mg.L-1, 0.2 ± 0.07 mg.L-1, and 0.2 ± 0.01 mg.L-1, respectively. These differences of the ammonium concentrations could be explained by the high densities of individuals in these breeding tanks particularly at the beginning of the experiment and the different efficiency of biological filters between tanks.

Indeed, a high density of individuals in the rearing tanks generates greater pollution of tank water, especially with the production of ammonium and nitrite. Willführ-Nast et al. (1993)[15] report that high levels of ammonium and nitrite negatively affect the larval growth. This could explain the larval growth delays noted in tanks C5, C7 and B5 where the ammonium levels were highest and exceeded the indicative value of 0.09 mg.L-1 recommended by Zweig et al. (1999) for the rearing of the species of the genus *Macrobrachium*. Tank C6, with an ammonium level of 0.09 ± 0.05 mg.L-1, also showed low larval growth despite the fact that the ammonium level was within the recommended level of ammonium. The time required for *M. vollenhovenii* to pass from larvae to post-larvae stage was relatively long compared with other prawns of the same genus. It took an average of 70 ± 10 days of production to move from zoea 1 to post-larvae stage, while for *Macrobrachium rosenbergii*, this time was estimated between 30 and 38 days on average (Nandlal *et al.,* 2005)[18]. A larval culture experiment on *M. vollenhovenii* was conducted by Coyle (2012) with gravid females from Cameroon who hatched them in a closed circulation culture system with a salinity of 16 ppt. He observed a very long larval period of between 65 and 75 days to obtain the first post-larval stage. Willführ-Nast et al. (1993) obtained the first PL stage of *M. vollenhovenii* in 45 days. In our six production tanks, B4, B5, C4, C5, C6 and C7, the first post-larval stages appeared on the 31st, 65th, 45th, 60th, 70th and 56th days after hatching, respectively. The duration of the larval phase (the elapsed time until the first post-larva stage) in tank C4 was comparable with that obtained by Willführ-Nast et al. (1993)[15]. The time in tanks B5, C5 and C6 was between the 65th and 70th day after hatching and was comparable to that obtained by Coyle (2012)[19].

On the other hand, in tank B4, the first PL stage was obtained 31 days after hatching. To our knowledge, no study has yet shown that *M. vollenhovenii* larvae can reach the post-larval stage 31 days after hatching. This performance could be explained by the water quality in this tank during the experiment. The lowest ammonium level was noted in tank B4 at 0.07 ± 0.04 mg.L-1. The time until the first PL stage noted in tank B4 was comparable to that obtained in the production of *M. rosenbergii*. This Asiatic species belong to the same genus of *M. vollenhovenii* and has been subjected to many studies around the world (Nandlal *et al.,* 2005; Barki *et al.,* 1991)[18-20]. *M. rosenbergii* has a short larval cycle with 11 stages, compared to 15 for *M. vollenhovenii*. Larval cultivation of *M. rosenbergii* takes 22 to 35 days on average (Nandlal *et al.,* 2005). In addition, larval growth stops were noted with *M. vollenhovenii*. The process of

metamorphosis seems to be blocked during the passage of the stages 4 to 5 and 9 to 10, as observed in tanks B5, C5, C6 and C7. The mortality during these passages was high. Makombu et al. (2014), in the three series of experiments carried out in Cameroon on *M. vollenhovenii*, noted the same phenomenon of blocked larval development between the 5 and 6 and between 9 and 10 stages along with a high larval mortality rate during these stages. These stages, marked by a blockage of the metamorphosis of *M. vollenhovenii* larvae, were also an origin of an asynchronous development of the larvae. From the hatching to the stage 5, larval growth was synchronous and rapid. After the stage 5, the development of the larvae was non-uniform in the tanks, except in tank B4. We noted up to four different larval zoea stages in tank C5. Nandlal et al. (2005)[18] reported the same phenomenon for *M. rosenbergii* bred in captivity, but the duration of larval blocking between the stages 5 and 6 was shorter than that observed for *M. vollenhovenii*.

Ultimately, the success rate of *M. vollenhovenii* larval production seem to be affected by three factors: the large number of larval zoea stages, 15 stages in total; the frequent stopping of larval metamorphosis in stages 4 to 5 and 9 to 10; and water quality in particular its ammonium and nitrite levels. Willführ-Nast et al. (1993)[15], working on the procedure and breeding conditions of *M. vollenhovenii* larvae, obtained a success rate of 12% of larvae metamorphosed into PL stage. The highest rate of successful larval production in this study was 3.08% obtained in tank B4 with better water quality and the lowest larvae density. The tanks C5, C6 and C4 follow with successful larval rates of 2.5%, 2.43% and 2.18%, respectively. The lowest metamorphosed rates of larvae into PL stage were observed in the tanks B5 and C7 with 1.5% and 0.17%, respectively. In this study, the average rate of post-larvae produced in all tanks was estimated at 2.8% of the total hatchery production. Apart from a success rate for PL production obtained by Willführ-Nast et al. (1993)[15], the average rate recorded in this study was similar or even better than those obtained by Coyle (2012)[19] with less than 2% and Makombu et al. (2014)[14] with 3% of PL stage production at *M. vollenhovenii*[21].

Conclusion

This study focused on the reproduction and larval rearing of *M. vollenhovenii* in the purpose to domesticate this species in a perspective of its use as a biological control agent against human schistosomiasis and to contribute to the development of aquaculture. It was intended to ensure the production and rearing of larvae from hatching to post-larvae production. The reproduction of broodstock was successfully done in fresh water at a water temperature of 28°C. The average number of post-larvae produced was estimated at 2.8% of the total larvae obtained. The first post-larvae were obtained on average 54 days after hatching. Water quality appeared to be one of the limiting factors in hatchery production of *M. vollenhovenii*. The control of the chemical parameters of water, such as ammonium and nitrite, seem essential to reduce the production cycle time and to improve the success rate of PL production of this species. The post-larvae coming from this hatchery production were grown in ponds and juveniles were transferred to the enclosures in selected villages with high prevalence rates of human schistosomiasis. This repopulation of the Senegal River ecosystem with *M. vollenhovenii* individuals produced in the hatchery aims to interrupt the transmission cycle of human schistosomiasis by biological control of mollusks, intermediate hosts of schistosomes and thereby contribute to the improvement of public health in the villages along the Senegal River.

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References

1. Sokolow, S.H., Lafferty, K.D., Kuris, A.M. Regulation of laboratory populations of snails (Biomphalaria and Bulinus spp.) by river prawns, Macrobrachium spp. (Decapoda, Palaemonidae): Implications for control of schistosomiasis. (2013) Acta Tropica 132: 64-67.
Pubmed | Crossref | Others
2. Swartz, S.J., DE Leo, G.A., Wood, C.L., et al. Infection with schistosome parasites in snail's leads to increased predation by prawns: implications for human schistosomiasis control. (2015) J Exp Biol 218: 3962-3967.
Pubmed | Crossref | Others
3. Savaya, A.A., Barki, O.O., Sagi, A. Size-selective predation by all-male prawns: implications for sustainable biocontrol of snail invasions. (2017) in Ben-Gurion University of the Negev, Israël. (2018) Biol Invasions 20: 137–149
Pubmed | Crossref | Others
4. Zabi, S. G. et Le Loeuff, P. La macrofaune benthique : 189-227. In : Environnement et ressources aquatiques de Côte d'Ivoire. (1994) II-Les milieux lagunaires (Durand,J. R., Dufour, P., Guiral, D., Zabi, S. G. F., eds). ORSTOM, Paris
Pubmed | Crossref | Others
5. Sokolow, S.H., Lafferty, K.D., Kuris, A.M. Regulation of laboratory populations of snails (Biomphalaria and Bulinus spp.) by river prawns, Macrobrachium spp. (Decapoda, Palaemonidae): Implications for control of schistosomiasis. (2013) Acta Tropica 132: 64-67.
Pubmed | Crossref | Others
6. Southgate, V.R. Schistosomiasis in the Senegal River Basin: before and after the construction of the dams at Diama, Senegal and Manantali, Mali and future prospects. (1997) J Helminthol 71(2): 125-132.
Pubmed | Crossref | Others
7. DeGeorges A. Reilly B. K. Dams and Large Scale Irrigation on the Senegal River. Impacts on Man and the Environment. (2006) Human Development, Report 2006/38, Human Development Report Office, Occasional Paper, 38.
Pubmed | Crossref | Others
8. Savaya-Alkalay, A., Rosen, O., Sokolow, S.H., et al. The prawn Macrobrachium vollenhovenii in the Senegal River basin: towards sustainable restocking of all male populations for biological control of schistosomiasis. (2014) PLoS Negl Trop Dis 8(8): e3060
Pubmed | Crossref | Others
9. Konan, K.M., Adépo-Gourène, A.B., Ouattara, A., et

al. Morphometric variation among male populations of freshwater shrimp Macrobrachium vollenhovenii Herklots, 1851 from Cote d'Ivoire Rivers. (2010) Fisheries Research 103: 1-8.
Pubmed | Crossref | Others

10. Bello-Olusoji, O.A. Assessment of the African River Prawn Macrobrachium vollenhovenii (Herklots, 1857) in some lentic and lotic environments in Nigeria. (1997) African Journal of Ecology 35(1): 80-87.
Pubmed | Crossref | Others
11. Anetekhai, M.A. Aspects of the bioecology of the African river prawn Macrobrachium vollenhovenii (Herklots) in Asejire Lake. (1986) Oyo State Nigeria.
Pubmed | Crossref | Others
12. Marioghae IE. Notes on the biology and distribution of Macrobrachium vollenhovenii and Macrobrachium macrobrachion in the Lagos lagoon (Crustacea, Decapoda, Palamonidae). (1982) Revue de Zoologie Africaine 94(3): 493–508.
Pubmed | Crossref | Others
13. Niass F., Fall, S. Reproduction and larval rearing of the freshwater prawn, Macrobrachium vollenhovenii in Senegal. (2016) International Journal of Biological and Chemical Science 9(6): 2523-2534
Pubmed | Crossref | Others
14. Makombu, J.G., Oben, P.M., Oben, B.O., et al. Complete Larval Development of the Fresh Water Prawn Macrobrachium vollenhovenii in Cameroon. (2014) Journal of Applied Aquaculture 26(4): 310-328.
Pubmed | Crossref | Others
15. Willführ-Nast, J., Rosenthal, H., Udo, P. J., et al.. Laboratory cultivation and experimental studies of salinity effects on larval development in the African river prawn, Macrobrachium vollenhovenii (Decapoda, Palaemonidae). (1993) Aquatic Living Resources 6: 115–137.
Pubmed | Crossref | Others
16. Anetekhai, M.A., Fagade, S.O. Induced spawning and laboratory rearing of the larvae of the African river prawn Macrobrachium vollenhovenii (Herklots) caught at Asejire Lake. (1987) Paper presented to the National Fisheries Development Committee meeting, Port Harcourt, Nigeria.
Pubmed | Crossref | Others
17. Zweig, R.D., Morton, J.D., Moral, M.S. Source Water Quality for Aquaculture: A Guide for Assessment. (1999) Washington D.C. Environmentally and socially sustainable development 76.
Pubmed | Crossref | Others
18. Nandlal, S., Pickering, T. Freshwater prawn Macrobrachium rosenbergii farming in Pacific Island countries. (2005) Volume one. Hatchery operation. Noumea, New Caledonia: 40.
Pubmed | Crossref | Others
19. Coyle, S.D. Freshwater Prawns: Biology and Farming. (2012) Aquaculture Rechearch Center: 544.
Pubmed | Crossref | Others
20. Barki, A., Karplus, I., Goren, M. The Agonistic Behavior of the 3 Male Morphotypes of the Fresh-Water Prawn Macrobrachium rosenbergii (Crustacea, Palaemonidae). (1991) Behaviour 116(3): 252-277.
Pubmed | Crossref | Others
21. FAO. Farming freshwater prawns. A manual for the culture of the giant river prawn (Macrobrachium roenbergii). (2002) FAO: Rome 428p.
Pubmed | Crossref | Others