Indian Journal of Geo-Marine Science
Vol. 40(3), June 2011, pp. 325-330

Environmental substrate selection and daily habitual activity in *Xiphopenaeus kroyeri* shrimp (Heller, 1862) (Crustacea: Penaeioidea)

Fulvio A M Freire [a], [b]Ana C Luchiari[c] & Vivian Fransozo[bd]

[a]Departamento de Botânica, Ecologia e Zoologia, Universidade Federal do Rio Grande do Norte – UFRN, Campus Lagoa Nova, Cx. P. 1524, cep. 59072-970, Natal, RN, Brazil.

[b]NEBECC (Núcleo de Estudos em Biologia, Ecologia e Cultivo de Crustáceos).

[c]Departamento de Fisiologia e Patologia, Universidade Federal do Rio Grande do Norte – UFRN, Campus Lagoa Nova, Cx. P. 1510, cep. 59078-970 Natal, RN, Brazil.

[bd]Instituto Federal de Educacâo, Ciencia e Technologia Baiano, Campus santalnes, Km 2.5 da BR420, Santalnes, Bahia, 45320-000, Brasil

[a][E-mail: fulvio@cb.ufrn.br]

Received 17 December 2008; revised 24 March 2010

Present study consists the study on environmental substrate preference and daily habitual activity of *X. kroyeri* shrimp. Six different substrate combinations were tested using silt and clay, various granulometries of sand, and gravel. Experimental tank for preference testing consisted of a 50 cm diameter cylindrical tank with 6 lateral compartments and a radial area in the middle. Every substrate combination prepared was randomly chosen for each compartment. Each substrate was spread to have a 5 cm thickness in the compartments. One isolated individual for each trial (n= 40) was used. Activities of the animals were observed 8 times a day, at 13:30, 14:30, 17:30, 18:30, 0:30, 1:30, 7:30 and 8:30 h. *X. kyoyeri* had a significant preference for very fine and fine sand. During the day light time burrowing activity was more intense while at night swimming was higher. Present results enabled us to conclude that *X. kroyeri* has preference for very fine and fine sand substrate.

[**Keywords**:. Shrimp, fishery, interstitial, aquaria, substrate]

Introduction

Preference tests have been a useful tool to study environmental conditions that may provide for the animal's welfare[1]. For instance, these tests focus on features which shrimps can distinguish, such as salinity[2], water temperature[3], pH[4] and food[5]. Food-preference tests were used to improve palatability of the diet[6], so that the more shrimp ingest, the more they gain weight. In fact, Ouellette *et al.*[7] observed that *Crangon septemspinosa* (Say, 1818) can improve foraging response on sandy substrate.

Marine benthic invertebrates, such as shrimp, substrate has been recognized as an influential driving factor in the survival of these organisms[8]. Substrate is not only their main source of food as they use to forage on it, but also their source of shelter, since shrimp burrow itself inside the substrate in order to avoid predation. Substrate characteristics can affect shrimp physiology and behaviour. One way to identify specific physical parameters that may improve shrimp fitness can be achieved by preference tests. Therefore, substrate preference tests can be a way to understand how the animal recognizes its environment and which substrate may represent the better place to live.

For shrimp species, which are able to burrow during the day and forage at night, such as *X. kroyeri*, suitable substrate can bring advantages for feeding[7], defense[8,9], or mating[8]. Substrate selection represents an important behaviour during settlement, because the "right choice" can have a deep impact on their fitness. According to Wahle & Steneck (1992)[10], the habitat selection may have a deep influence on the pattern of distribution and abundance of early benthic stages.

Several studies have demonstrated substrate influence on some shrimp biological processes, since shrimps live on the substrate or are buried in it[11]. Kenion *et al.*[12] showed that fine sand is better cover for the *Penaeus esculentus* (Haswell, 1879) as a way to prevent predation. These organisms are subjected

corresponding author[a]

to high predation pressures from large benthic invertebrates and demersal fishes[10] and, time and again this interaction result in a high mortality rate. Hence, if a different substrate used by individuals provides differential protection against predators, then spatial distribution would be a reflection of substrate specific harmful effect. In this case, predation would exert a strong direct influence on the animal's substrate preference. Indeed, the substrate composition can affect shrimp survival[13] and growth rate[14], and it can be directly related to the time shrimp spend buried or out of the sediment[15].

Present study consists the environmental substrate preference and also burying and swimming activities of *X. kroyeri* shrimp during the light/dark cycle. We compared six different combinations of substrates, as marine aquatic environment is characterized by a wide variety of bottom substrates, where we can find from gravel to silt and clay[7]. It was hypothesized that *X. kroyeri* shrimp would prefer the substrate where it can burrow and forage with less energy costs. Knowledge of *X. kroyeri* preferred environmental substrate selection will lead to a better understanding of its ecology and distribution patterns in natural habitat.

Materials and Methods

Specimens of *X. kroyeri*, from both sexes, different ages and sizes, were sampled at Ubatuba region, State of São Paulo (23° 26' S and 45° 02' W), Brazil. Shrimp were captured by a commercial fishery boat supplied with 2.5 m opening double-rig trawling nets. The mesh size was 15 mm except in the cod, where it was 10 mm. Individuals were collected during the morning period (7:00 am) at 5 m depth. After that, shrimp were put in a thermal box containing seawater (27ºC; 35‰ salinity) and air supply. At the laboratory, in Ubatuba city, shrimps were transferred to a stock tank where they stayed for 6 hours until the beginning of the experimental procedures. During this period, oxygen saturated sea water was kept at 27 ºC average temperature and 35‰ salinity. Natural light conditions, prevailing around 12 h light and 12 h dark during spring time in Ubatuba, SP were used. Sampling and experimental procedures were performed in October 2004 (spring).

Substrates used in this study were prepared from natural environmental sediment collected in beach regions. Fine and very fine sand were collected from the supralittoral zone of Grande beach, and very thick, thick and medium sand were collected from supralittoral zone of Vermelha do Norte beach, both located in Ubatuba region. Silt and clay were collected from the supralitoral zone of Jabaquara beach, located in Parati-Rio de Janeiro. All substrates were sifted in different net sieves, so that it could be separated by increasing size. After sifting, substrates were washed in fresh water and dried under the sun. Substrates were grouped after the granulometry classification from Wentworth[16]: substrate *a* -composed by medium, thick, very thick sand with gravel (0.25 to > 2.0 mm), substrate *b* - composed by very fine and fine sand (0.0625 to 0.25 mm), and substrate *c* - composed by silt and clay (< 0.0625 mm).

Six different grain size substrate combinations were used in this study: *a*, *a*+*b*, *b*, *a*+*c*, *b*+*c* and *c*. After sifting and drying, substrate *c* was also burned in a semi-industrial oven (400ºC) to remove organic matter. For substrates *b* and *a*, a wash and dry was enough to remove the organic matter.

The experimental tank for preference test consisted of a 50 cm diameter cylindrical tank with 6 lateral compartments and a radial area at the middle (Fig. 1). Each lateral compartment had identical area and was equally illuminated by natural daylight (12L:12D). Different substrates were randomly chosen for each

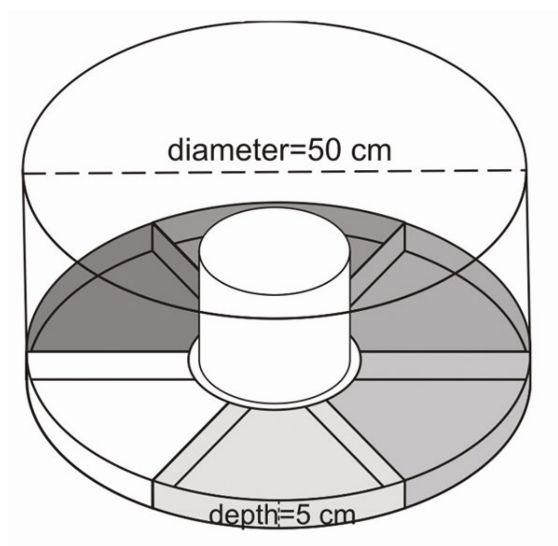

Figure 1. Diagram of the experimental tank used to test substrate preference of shrimp *Xiphopenaeus kroyeri*. The tank was 50 cm in diameter, divided into six radial compartments around a central area. The substrate for each compartment was obtained by the combinations of silt and clay, various granulometries of sand, and gravel. Each substrate was spread to have 5 cm thickness in the compartments. The central area did not receive any substrate. Seawater (35‰ salinity) was supplied until 20 cm of the bottom of the tank. The tank was under natural light conditions.

compartment. Each substrate with 5 cm thickness was used in the compartments. The central area where the shrimp had been introduced was not supplied with substrate in order to prevent shrimp from being there. Tanks were supplied with seawater, as used to keep shrimp in the stock tank.

Forty shrimps isolated in experimental tank (1shrimp/tank) were initially placed in a white tube in the middle area for 30 minutes, at 13:00 o'clock. After this period, the tube was removed and "visit frequency" at each different substrate was observed. Each shrimp was observed 8 times during a 24 -hour period. Shrimp visit frequency at each compartment was observed every 2 minutes for 20 minutes during day (at 13:30 and 14:30 h), dusk (at 19:30 and 20:30 h), night (at 00:30 and 01:30 h) and dawn (at 05:30 and 06:30 h). At each observation period, it was also registered if the animal was swimming, buried or placed on the substrate and at which substrate the shrimp was located at. During the dark period, shrimps were observed under dim red illumination (610 nm, 15lx) to avoid light interference.

For statistical analysis, the non-parametric procedure of Friedman Anova was used for multiple group analyses since all attempts to obtain a normal distribution have failed. We also used the Friedman test because shrimp preference for one substrate instead of others provides dependent data. In cases where the Friedman test was significant ($\propto$<0.05), data was compared by Student-Newman-Keuls post hoc test[17] to determine significant differences among substrates.

Results

Mean visit frequency of shrimp in each substrate is shown in Fig. 2. Significant preference of *X. kroyeri* was observed for very fine and fine sand, substrate *b* (Friedman test, χ^2=488.9 e p=2.09×10^{-102}) instead of the other portions. Preference test results showed that *X. kroyeri* has first selected very fine and fine sand as most preferred substrate, followed by silt and clay, and the last preferred substrate was very thick, thick and medium sand with gravel.

Burying behaviour was dependent on daily light cycle. Burying activity was higher during the light (day) than during dark (night) periods (Friedman test, χ^2=100.3 and p=9.47×10^{-19}), as it is shown in Fig. 3. At dawn and dusk, burying and swimming activities were intermediary between their behaviour at night and day periods. During the night period, we could observe high swimming activity, as shown in Fig. 4.

We also observed that water became a bit turbid when the animals began to bury in the silt and clay substrate. When shrimps tried to bury in silt and clay,

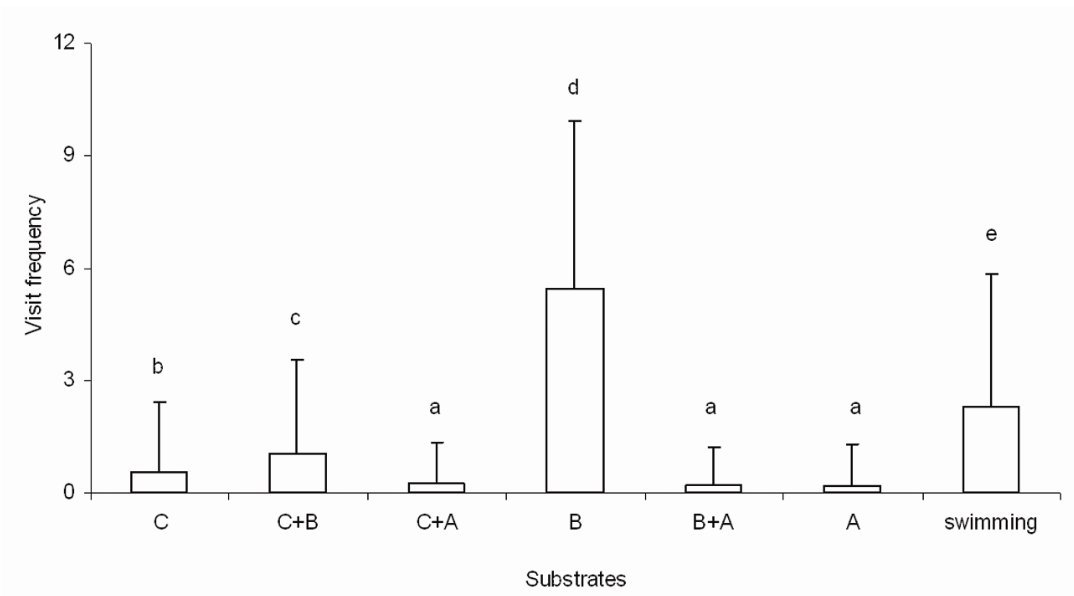

Figure 2. Shrimp *Xiphopenaeus kroyeri* visit frequency in each substrate in a preference test and swimming activity (± SD) N=40. Bar values are the means of eight observation periods (13:30, 14:30, 19:30, 20:30, 0:30, 1:30, 5:30 and 6:30 h) during 24 h period. Substrate tested were c = silt and clay; c+b = silt and clay + thin and very thin sand; c+a = silt and clay + thick, very thick and medium sand + gravel; b = thin and very thin sand; b+a = thin and very thin sand + thick, very thick and medium sand + gravel; a = thick, very thick and medium sand + gravel. Different letters indicate statistically differences at $p < 0.05$ (Friedman ANOVA followed by Newman-Kuels test).

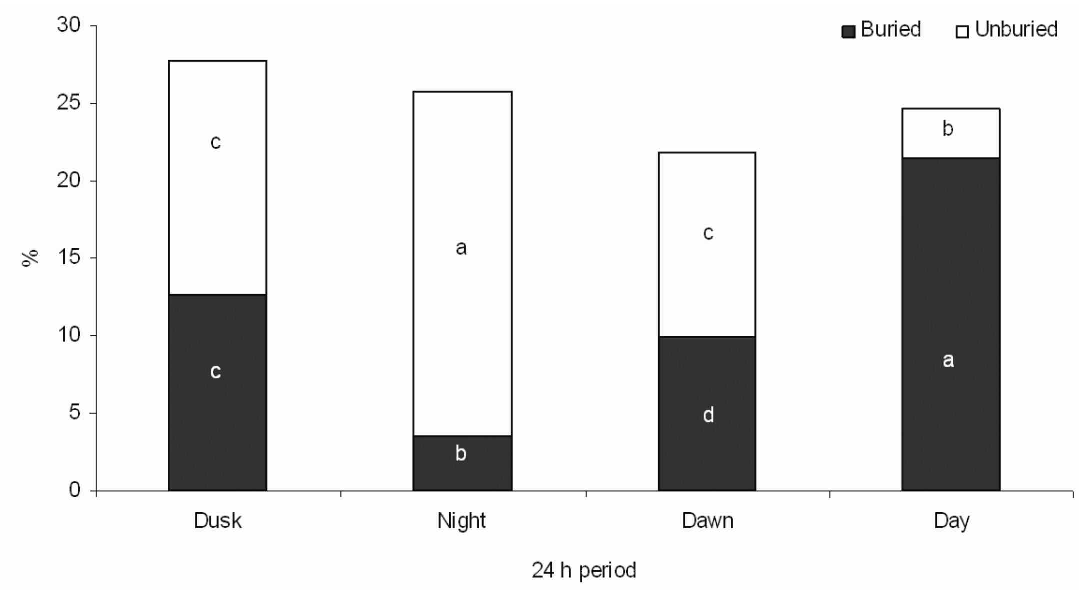

Figure 3. Shrimp *Xiphopenaeus kroyeri* buried and unburied time percentage during four different periods of the day cycle. Different letter indicate statistical differences at $p< 0.05$, N=40 (Friedman ANOVA followed by Newman-Keuls test).

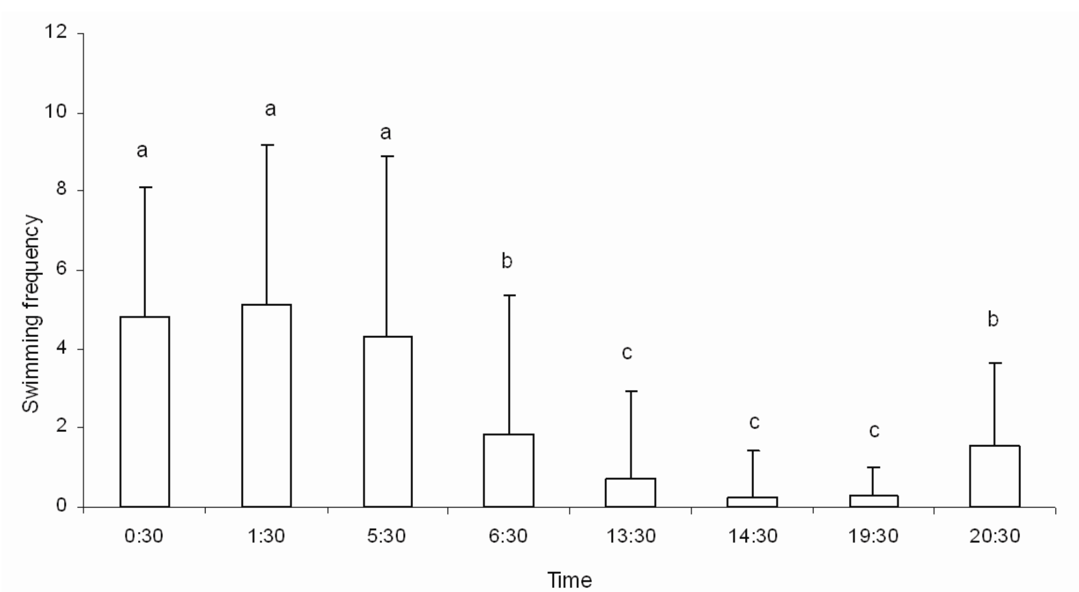

Figure 4. Shrimp *Xiphopenaeus kroyeri* swimming activity (± SD) N=40. Bar values are the means of eight observation periods (13:30, 14:30, 19:30, 20:30, 0:30, 1:30, 5:30 and 6:30 h) during 24 h period. Different letter indicate statistical differences at $p< 0.05$ (Friedman ANOVA followed by Newman-Keuls test).

sediment particles scattered in the water. Hence, shrimp were inside a hole in the substrate but not covered.

Discussion

X. kroyeri shrimp preferred very fine and fine sand, it has markedly bury behaviour during the day and emerges out of the substrate at night. Accordingly to Dall *et al.*[8], the majority of peneid shrimp has preference for sediment having particles size between 62 µm and 1 mm and it occurs due to burying facilities and ability to breathe when buried in sand substrate.

The *X. kroyeri* preference for very fine and fine sand can be explained by the ease of excavation that this granulometric substrate allows. Marine shrimp uses excavation as a hiding strategy against predators, thus, the substrate must be favourable for fast burying and hiding. Other shrimp, as *Penaeus sculentus*, *Penaeus semisulcatus* de Haan 1844 and *Penaeus monodon* Fabricius 1798, also have this strategy to protect themselves and they show preference for fine sand[9,12]. According to Day[18], substrates consisting of particles around 2 mm diameter have higher erosion tendency, hence, it is easier and faster for shrimp to bury in very fine and fine sand.

In substrates where it is difficult to bury, their capture by the predators is increased because shrimp try to excavate but the substrate does not cover their bodies well and they are easily found[19]. Ouellette *et al.*[7] suggested that shrimp burying behaviour is less effective in peat than in sand substrate, since peat particles scatter in the water due to its low haste capacity. Thus, extremely fine substrates, as silt, clay and peat, do not haste just after being touched, leaving the animal partially exposed.

Furthermore, silt and clay substrate present decreased interstitial spaces, and thus low oxygen level, which may affect oxidative nitrification reaction when in the presence of organic matter. In cases where organic material is very highly concentrated, anaerobic processes take place and toxic metabolic substances are released[20]. Allan & Maguirre[15] found high ammonia concentration in aquaria with silt and clay substrate, possibly due to high organic matter condition and little interstitial space for oxygen presence. In this condition, shrimp emerged from substrate during light periods probably because of the ammonia level. Hence, silt and clay geomorphology may contribute to a decrease in water and soil quality.

On the other hand, thick substrates have big particles, which are not easily moved and only big shrimp species, as *Farfantepenaeus aztecus* (Ives 1891) and *Penaeus setiferus* (Linnaeus 1767), showed preference for very thick sand substrate[21]. Since shrimp studied herein (*X. kroyeri*) belong to a medium sized species, they are able to bury without difficulty in very fine and fine sand, as other species of the same size, for example *Crangon septemspinosa*[7].

According to Dall *et al.*[8], fine sand also has clay and water in its composition, which makes this substrate more fluid and improves burying activity. For this author, shrimp preference for fine sand substrate is affected by particle size distribution, interstitial space and porosity among particles, water content and organic matter presence. Other studies have already addressed the improved feeding behavior that shrimp can experience when foraging on sand substrate, which offers organic matter. For instance, *Penaeus esculentus* (Ruello 1973), *Penaeus japonicus* (Bate 1888) and *Penaeus kerathurus* (Forskal 1775)[22] have higher foraging and growth performance in sand substrate.

Although fine sand preference can be affected by the presence of organic matter, in this study substrate preference could not address the fact since all substrate used were cleaned to avoid organic material. This result is in agreement with the conclusion of Williams[23], that food availability is not an important factor on peneid substrate preference because these animals' choice is not related to presence or absence of food. Therefore, fine sand preference should be explained by other factors, such as burying and breathing ease.

During the breathing process, shrimp gills receive water influx directed by the respiratory tube, which is composed by anterior appendices. After that, water is expelled from the gill chamber beneath its carapace. When shrimp are buried, expiration process causes substrate particles to percolate to the substrate surface. Placed inside extremely fine substrate (silt and clay), the particles' reverberation during expiration can obstruct the gills chamber and, thus, breathing is facilitated under thicker substrate particles, as sand[8].

Beside *X. kroyeri* the thin sand preference already discussed above, we could also observe the effects of day cycle on its activity. It is well known that penaeid shrimp bury during the day light period and emerge to forage and swim at night[21,23]. A determinant factor affecting this behaviour is light incidence period. Light may inhibit shrimp emerging behaviour, and factors affecting luminosity are related to this animals capture levels, as seasonal cycle, moon phase and water turbidity[24]. Moller & Jones[9] and Sanches[25] described increased shrimp activity during the night and Carothers & Chittenden[26] observed higher capture when environmental conditions decrease light availability, for example after storms. This kind of behavior is also related to predator avoidance, since it is decreased at low visibility periods.

While buried, shrimp are inactive and its metabolic rate is very low[27]. As observed herein, during the light period shrimp was mainly buried and they got out of the substrate at night. During the dark periods, high swimming activity was noticed, which emphasizes shrimp's nocturnal behaviour. Moreover, high predation stress on shrimp may have molded this animal's ability to perceive light and bury as a way to protect itself. In addition, to be on thin sand may improve burying ability and allow good breathing condition for the hiding animals.

Therefore, we can conclude that *X. kroyeri* has preference for thin sand substrate and it is a markedly nocturnal behaviour animal. These characteristics corroborate to what have already been observed for the majority of Penaeidae species. This data is also important to increase knowledge of optimal peneid shrimp cultivation in captivity and to improve penaeid shrimp fishing efforts, as the space and temporal distribution of the species can be estimated by substrate type and daytime.

Acknowledgement

Authours are grateful to the "Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP)" (Grant # 94/4878-8, # 98/03134-6 and # 02/02274-6) for financial support.

References

1 Gonyou, H.W., Why the study of animal behaviour is associated with the animal welfare issue, *J. Anim. Sci.*, 72(1994) 2171-2177
2 Bray, W.A., Lawrence, A. L., Effect of four substrates on growth and survival of *Penaeus vannamei*, *Cienc. Mar.*, 19(1993) 229-244
3 Reynolds, W.W., Casterlin, M.E., Thermoregulatory behavior of the pink shrimp *Penaeus duorarum* Burkenroad, *Hydrobiologia*, 67(1979) 179-182
4 Lemonnier, H., Bernard, E., Boglio, E., Goarant, C., Cochard, J., Influence of sediment characteristics on shrimp physiology: pH as principal effect, *Aquaculture*, 240(2004) 297-312
5 Omori, M., Preliminary rearing experiments on the larvae of *Sergestes lucens* (Penaedia, Natantia, Decapoda), *Mar. Biol.*, 9(1971) 228-234
6 Fraser, D., Assessing animal well-being: common sense, uncommon science. *Food Animal Well-Being,* (1993) 37-54
7 Ouellette, C., Boghen, A.D., Courtenay, S.C., St-Hilairem, A., Influence of peat substrate on the distribution and behaviour patterns of sand shrimp, *Crangon septemspinosa*, under experimental conditions, *J. Appl. Ichthyol.*, 19(2003) 359-365
8 Dall W, Hill B J, Rothlisberg P C & Sharples D J, *The biology of the Penaeidae. Advances in Marine Biology* (Academic Press, San Diego) 1990, pp. 489
9 Moller, T.H., Jones, D.A., Locomotory rhythms and burrowing habits of *Penaeus semisulcatus* (de Haan) and *P. monodon* (Fabricius) (Crustacea: Penaeidae), *J. Exp. Mar. Biol. Ecol.*, 18(1975) 61-77
10 Wahle, R.A., Steneck, R.S., Habitat restrictions in early benthic life experiments on habitat selection and in site predation with the American lobster. *J. Exp. Mar. Biol. Ecol.*, 157(1992) 91-114
11 Fuss, C.M., Ogren, L.H., Factors affecting activity and burrowing habits of the pink shrimp, *Penaeus duorarum* Burkenroad, *Biol. Bull.*, 130(1966) 170-191
12 Kenyon, R.A., Loneragan, N.R., Hughs, J.M., Habitat type and light affect sheltering behaviour of juvenile tiger prawns (*Penaeus esculentus* Haswell) and success rates of their fish predators, *J. Exp. Mar. Biol. Ecol.*, 192(1995) 87-105
13 Méndez, L., Racotta, I., Acosta, B. Portillo-Clark, G., Effect of sediment on growth and survival of postlarval *Litopenaeus stylirostris* (Boone, 1931), *Aquacult. Res.*, 35(2004) 1-7
14 Bratvold, D., Browdy, C.L., Effect of sand sediment and vertical surfaces (AquaMatsþ) on production, water quality and microbial ecology in an intensive *Litopenaeus vannamei* culture system, *Aquaculture*, 195(2001) 81-94
15 Allan, G.L., Maguire, G.B., Effect of sediment on growth and acute ammonia toxicity for the school prawn, *Metapenaeus macleayi* (Haswell), *Aquaculture*, 131(1995) 59-71
16 Wentworth, C.K., A scale of grade and terms for cladistic sediments, *J. Geol.*, 30(1922) 377-392
17 Zar J H, *Biostatistical Analysis,* (Prentice Hall, Engelwood Cliffs, New Jersey) 1999, pp. 663
18 Day J H, Coastal hydrodynamics, sediment transport and inlet stability, in *Estuarine Ecology,* (Blakema, Rotterdam) 1981, pp. 7-25
19 Minello, T.J., Zimmerman, R.J., Martinez, E.X., Fish predation on juvenile brown shrimp, *Penaeus aztecus* Ives: effects of turbidity and substratum on predation rates, *Fish. Bull.*, 85(1987) 59-70
20 Boyd, C.E., *Bottom Soils, Sediment, and Pond Aquaculture* (Chapman and Hall, New York) 1995, pp. 348
21 Yip-Hoi, T.A., *An Investigation of Effects of Dissolved Oxygen Level, Sediment Type, Stocking Density and Predation on the Growth Rate, Survivorship, and Burrowing Behavior of Juvenile Brown and White Shrimp*, Ph.D. thesis, North Carolina State University, USA, 2003
22 Otazu-Abril, M., Ceccadi, H.J., Contribution a l'etude du comportement de *Penaeus japonicus* (Crustacea Decapode) en elevage, vis-a-vis de la lumiere et du sediment, *Tethys*, 10(1981) 149-156
23 Williams, A.B., Substrates as a factor in shrimp distribution, *Limnol. Oceanogr.*, 3(1958) 283-290
24 Penn J W, *Penaeid Shrimps – Their Biology and Management*, (Fishing New Books, Farnham, England) 1984, pp. 308
25 Sanchez, A.J., Habitat preference of *Penaeus duorarum*, Burkenroad (Crustacea: Decapoda) in a tropical coastal lagoon, southwest Gulf of México, *J. Exp. Mar. Biol. Ecol.*, 217(1997) 107-117
26 Carothers, P.E., Chittenden, M.E., Relationship between trawl catches and tow duration for penaeid shrimp, *T. Am. Fish. Soc.*, 114(1985) 851-856
27 Dali, W., Estimation of routine metabolic rate in a penaeid prawn, *Penaeus esculentus.* Haswell, *J. Exp. Mar. Biol. Ecol.*, 96(1986) 57-74