

## Size of suspended particles caught by Manila clam, *Ruditapes philippinarum*

Hisayuki ARAKAWA\*, Toshihide YAOITA\*, Takashi KOIKE\*\*  
and Tsutomu MORINAGA\*\*

**Abstract:** The sizes of suspended particles caught by common Manila clam and the shell-length variations in the selection of particle sizes were investigated using a Coulter Counter. A total of 450 samples were examined under 4 different shell lengths, viz. 5, 10, 20, and 30mm. The simulated feed suspensions were either the phytoplankton *Pavlova lutheri* (average size 3.5  $\mu\text{m}$ ) or pulverized pellet powder (average size 3.7  $\mu\text{m}$ ), which is used as artificial feed for abalone. The percent catch rate was based on the proportion of particles reduced from the initial number. The particle sizes examined were in the range 2.2 to 45.2  $\mu\text{m}$ .

The catch-rates of adult clam (shell length of 30mm) under vigorous filtration indicated nearly constant particle size selection when they were maintained for 60 minutes in the sea water containing  $3 \times 10^5$  pellet particles/ml. When the particle concentration increased to  $6 \times 10^5$ /ml, however, the catch rates of sizes less than 15  $\mu\text{m}$  decreased to almost a third of the above mentioned concentration. A similar reduction was noted when the exposure was for 180 minutes. Furthermore, the catch rates for particle sizes more than 5  $\mu\text{m}$  increased by almost 20% when pellet was replaced with phytoplankton.

When Manila clam of different shell lengths were kept for 60 minutes in the sea water containing  $3 \times 10^5$  particles/ml, the catch rates of 5 mm clams for the entire range of particle sizes were almost uniform, showing an average value of  $0.68\% \cdot \text{h}^{-1}$ . When their shell lengths increased to 10 mm or 20mm, the catch rates for the larger sizes appeared to increase. Thus the larger clams, among the sizes examined, seeks intentionally larger suspended particles to meet their growth requirements.

### 1. Introduction

The bivalve Manila clam, *Ruditapes philippinarum*, is found abundantly in the coastal waters all around Japan. Since it feeds on suspended particles, it is considered as one of the important benthos from the view-point of cleaning or purifying sea water (AKIYAMA, 1985; AOYAMA and SUZUKI, 1997). The growth of the clam's natural larvae as well as the artificially introduced seed-shells is influenced by not only such environmental factors as coastal water properties, bottom materials, and preda-

tors but also their feeding itself. Particularly, the latter is considered to exert the most serious influence on the clam's growth in their early stages.

A great deal of information exists on the feed and filtration of bivalves. For instance, those of oyster reported by LOOSANOFF and ENGLE (1947), KUSUNOKI (1977 a, b, c) and RIISGÅRD (1988). Information on sea mussel has been provided by JØRGENSEN (1949), UMEZU *et al.* (1967) and LUCAS *et al.* (1987) and others. These authors investigated the quantities of suspended particles caught by oyster and mussel according to particle sizes.

With regard to studies on Hard clam and Manila clam, there are the reports of CHIBA and OHSHIMA (1957), FURUKAWA (1961), NUMAGUCHI

\* Department of Ocean Sciences, Tokyo University of Fisheries, 5-7, Konan 4, Minato-ku, Tokyo 108, Japan

\*\* Faculty of Bioresources, Mie University, Kamihama-cho, Tsu-shi, Mie 514, Japan

(1990), TOBA and MIYAMA(1993) and MUKAI (1993). CHIBA and OHSHIMA(1957) investigated the influence of muddy water on the filtration process of several bivalves including Manila clam, and reported that their rates did not go down even in the water suspended thickly with bentonite particles. Recently, NUMAGUCHI (1990) investigated the distributions of Manila clam and the properties of suspended particles around the river mouth in the Bay of Ariake, and suggested that the particles selected as feed were in the size range from 1.2 to 50  $\mu\text{m}$  and richly pigmented.

Apart from the work of TOBA and MIYAMA (1993), there is very little information on the size-preferences in selection of suspended particles by Manila clam, let alone the shell length based selection. Therefore this study broadly attempts to examine (1) the change of particle catch-rates with time, (2) the quantity and quality of those particles depending on filtration performance, and (3) to find out the sizes of captured-particles in relation to the different shell-lengths of the clam.

## 2. Materials and methods

### 2-1. Samples and materials

Artificial seed-shells (shell length : 5 and 10 mm) produced at Futtsu Branch of Chiba Prefectural Fisheries Experimental Station, as well as natural clams (shell length : 20 and 30 mm) collected from the Tokyo Bay were used as the samples. A total of 450 clams were used for the experiments and the observational error in shell length was restricted to be less than  $\pm 0.1$  mm. Each sample was kept from one to three months in basin with filtered sea water maintained at 21°C.

The feed particle suspensions were either artificial feed or phytoplankton. The artificial feed employed in this experiment was a commercial pellet manufactured by Halios Japan Combination Feed Co. Ltd, for abalone. The pellet was finely ground and the resulting powder, was mixed with sea water filtered through millipore CP15 (pore-size 1  $\mu\text{m}$ ), and later filtering the suspension itself through a net of mesh size 50  $\mu\text{m}$ , the average size of the resulting particle suspension being 3.7  $\mu\text{m}$ . On the other hand, the phytoplankton *Pavlova lutheri*

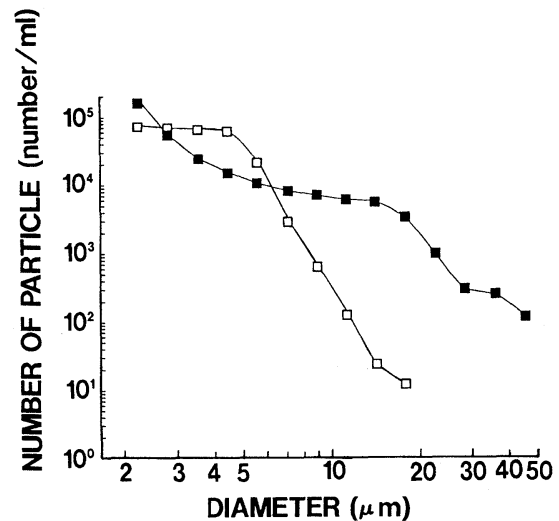


Fig. 1. The particle size distributions of the pellet and phytoplankton offered to the Manila clam.

Symbols  $\blacksquare$  and  $\square$  denote the particle sizes of artificial feed (pellet) and phytoplankton *Pavlova lutheri*, respectively.

was monocultured in a constant temperature room, and was allowed to emigrate into the sea water of the experimental basin, at the average size of 3.5  $\mu\text{m}$ . The phytoplankton culture was considered to include both clastic cells and agglutinative forms. The particle size distributions of pulverized pellet and phytoplankton are shown in Fig. 1.

### 2-2. Experimental methods

The experimental apparatus is illustrated in Fig. 2. The experimental tank was cylindrical in shape, and had a diameter of 15 cm and depth of 10 cm. This tank contained yet another cylindrical-shaped inner container, 4 cm both in dia. and depth, which held the bivalves. A stirrer was provided to facilitate water circulation.

The animals were deprived of food for 24 hours prior to the start of a series of experiments. The number of samples under the shell-lengths of 5 and 10 mm were forty and twenty respectively, whereas for the 20 and 30 mm size only individual clams were used. Artificial feed particles were added to one liter of filtered sea water so that it produced suspensions with concentration of  $1 \times 10^5$ ,  $3 \times 10^5$ , and  $6 \times 10^5$  part-

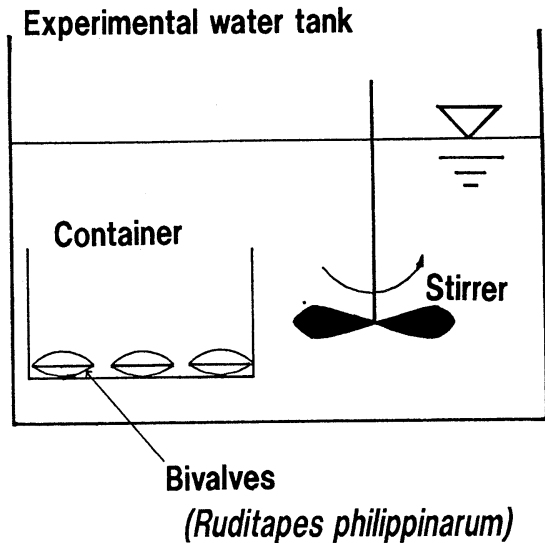


Fig. 2. Diagrammatic representation of the experimental set up.

icles/ml while the test phytoplankton concentration was  $3 \times 10^5$  cells/ml. After leaving the sample clam for 30, 60 or 180 minutes in the sea water at the particle concentrations mentioned above, the clam's retainer on the bottom of the vessel was taken out in order to avoid a possible contamination of the water with the clam's excrement.

The number of particles in the vessel was measured with Coulter Counter Model ZM (aperture size of  $100 \mu\text{m}$ ). The objective particles for the study were those in the range from 2.2 to  $45.2 \mu\text{m}$ . A control was employed simultaneously using the vessel without the experimental animals, and then the number of particles were counted.

Almost soon after each clam was let into the experimental container, it extended its incurrent siphon and started vigorous filtration. The clam used in the present experiment performed active filtration with the end of the incurrent siphon opened (MUKAI, 1993).

The catch rate,  $Cr_{(i)}$ , is obtained using the following formula :

$$Cr_{(i)} = \frac{1}{n \cdot t} \frac{Cc_{(i)} - Ce_{(i)}}{Cc_{(i)}} \cdot 100 \quad (1)$$

where,  $Cc_{(i)}$  is the concentration of particles without the clam,  $Ce_{(i)}$  denotes the concent-

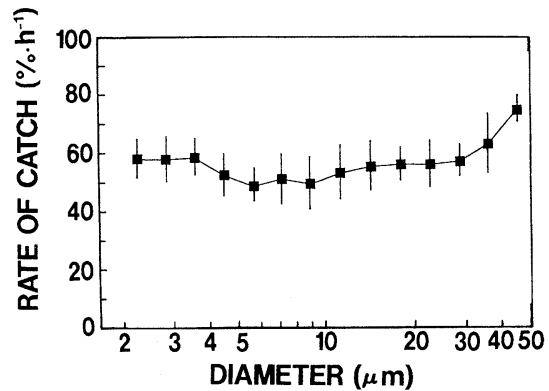


Fig. 3. Particle catch rates of adult Manila clam. The solid squares are average values from 11 clams, along with standard deviations.

ration of particles (diameter at  $i$ -class :  $di$ ) contained in the water with the clam,  $t$ (hour) stands for the experimental period, and  $n$  is the number of clams. Besides, the average diameter of captured particle ( $\bar{D}$ ) is based on the following formula :

$$\bar{D} = \frac{\sum di \cdot Ndi}{\sum Ndi} \quad (2)$$

Where,  $di$  stands for the diameter of particle-size at  $i$ -class, and  $Ndi$  stands for the number of particles at the diameter  $di$ .

### 3. Results

#### 3-1. Particle Catch-Rates of Adult Clam

##### 3-1-1. Changes with time

Figure 3 depicts the particle catch rate of the clam in a state of motion, when it was kept for 60 minutes in sea water containing  $3 \times 10^5$  pellet particles/ml. The catch rates were  $57.8\% \cdot \text{h}^{-1}$  corresponding to the particle size of  $2.8 \mu\text{m}$ ,  $53.1\% \cdot \text{h}^{-1}$  for  $11.3 \mu\text{m}$  size, and  $63.2\% \cdot \text{h}^{-1}$  for  $35.9 \mu\text{m}$ . Speaking generally, they exhibited catch rates within the range  $50$  to  $60\% \cdot \text{h}^{-1}$  for particle size less than  $25 \mu\text{m}$ , but slightly higher for those larger than that. Besides, the total volume of particles caught by adult was  $0.031 \text{ mm}^3$ . From these results it could be concluded that when the clam is in a state of motion, the particle catch rate distributions are almost uniform over a wide range of particle sizes, the median value being approximately  $60\% \cdot \text{h}^{-1}$ .

Figure 4 depicts the change catch rates in relation to the particle sizes, and the length of

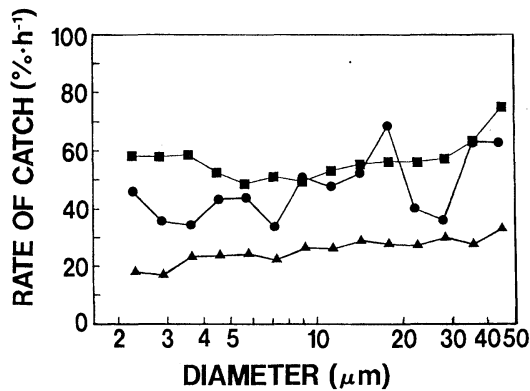


Fig. 4. Changes in the catch rates of adult clams under varying experimental periods.

Symbols ●, ■, and ▲ represent the rates for periods of 30, 60, and 180 minutes, respectively. Values are means of 6, 11, and 2 observations for periods of 30, 60, and 180 minutes.

exposure time. The experimental clams were introduced into the sea water of  $3 \times 10^5$  particles/ml, for durations of 30, 60, and 180 minutes. The data for 60 minutes exposure is the same as that in Fig. 3. The particle catch rates measured after being submerged for 30 minutes were  $35.6\% \cdot h^{-1}$ ,  $47.0\% \cdot h^{-1}$ , and  $62.6\% \cdot h^{-1}$  corresponding to particle sizes of  $2.8 \mu m$ ,  $11.3 \mu m$ , and  $35.9 \mu m$ , respectively. These rates after being left for 60 minutes were  $57.8\% \cdot h^{-1}$ ,  $53.1\% \cdot h^{-1}$  and  $63.2\% \cdot h^{-1}$ , respectively; and after 180 minutes of submergence the values were  $17.0\% \cdot h^{-1}$ ,  $25.3\% \cdot h^{-1}$ , and  $26.7\% \cdot h^{-1}$ , respectively. Thus, the average particle catch-rate to the whole range of the particle sizes varied widely when the clam was submerged for only 30 minutes. However there was uniformity in the capture rates for the different size groups when the clams were submerged for 60 minutes (about  $60\% \cdot h^{-1}$ ), and 180 minutes (only around  $20\% \cdot h^{-1}$ ). The variation in the catch-rates for the group exposed for the shortest duration lacks a proper explanation.

### 3-1-2. Changes with particle quality and quantity

In order to investigate the influence of particle concentration on catch rates, adult clams were treated as in the earlier description. Fig. 5 shows the particle catch rates classified by different particle concentrations. The results

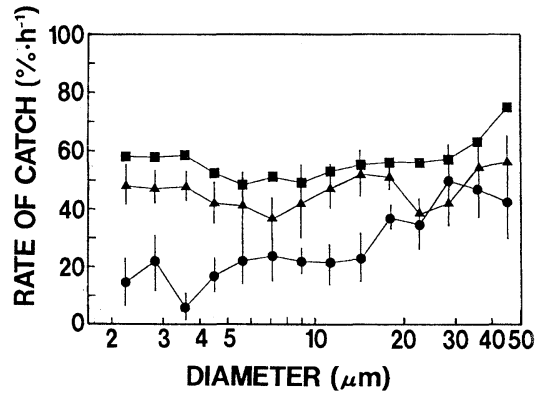


Fig. 5. Changes in the catch rates of adult clams under different concentrations of the suspended particles.

Symbols ▲, ■, and ● denote concentrations of  $1 \times 10^5$ ,  $3 \times 10^5$ , and  $6 \times 10^5$  particles/ml, respectively. Values are means of 3, 11, and 5 observations for the concentrations  $1 \times 10^5$ ,  $3 \times 10^5$ , and  $6 \times 10^5$  particles/ml, along with their standard deviations.

under the conditions of  $3 \times 10^5$  particles/ml exhibited in Fig. 5 are the same as those introduced in Fig. 3. The catch rates for the range of particle-sizes tested were approximately 40 to  $50\% \cdot h^{-1}$  for the particle concentration of  $1 \times 10^5$  particles/ml, and around  $60\% \cdot h^{-1}$  for the higher concentration of  $3 \times 10^5$  particles/ml. However, at the highest particle concentration of  $6 \times 10^5$  particles/ml, the catch rates were greater for the large size particles. The values were  $21.2\% \cdot h^{-1}$  and  $42.0\% \cdot h^{-1}$  for the particle-sizes  $14.2 \mu m$  and  $45.2 \mu m$ , respectively. On the other hand, when the particle assimilation was expressed on a volume basis, the ratios were 1.0 : 4.4 : 5.0 for the corresponding concentrations of  $1 \times 10^5$ ,  $3 \times 10^5$ , and  $6 \times 10^5$  particles/ml. This is a clear indication that the sample clam prefers larger particles.

Figure 6 shows the phytoplankton catch-rates corresponding to the different cell-sizes after allowing for 60 minutes in the test medium. The results were compared with the identical data of the pellet particle catch-rates exhibited in Fig. 3. As mentioned earlier, the catch-rates of the pellets was approximately  $60\% \cdot h^{-1}$ , irrespective of the particle sizes. The phytoplankton cell catch rates, however, was

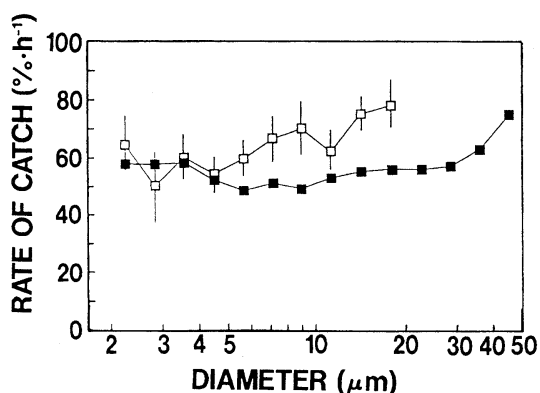


Fig. 6. Changes in the catch rates of adult clams when offered two different foods.

Symbols ■ and □ represent pellet and phytoplankton, respectively.

The corresponding sample numbers were 11 and 3 for pellet and phytoplankton. Standard deviations of the values are also indicated.

$50.3\% \cdot h^{-1}$  for the cell-size of  $2.8 \mu m$  and  $62.3\% \cdot h^{-1}$  for  $11.3 \mu m$  size, indicating increased rates for larger cells. It was noted that for both phytoplankton and feed pellets, the catch rates were uniform for particle sizes less than  $5 \mu m$ , but above this size the catch rates of phytoplankton increased by approximately 20%. This result suggests that the mollusc tends to discriminate the particle quality when the particles are larger than  $5 \mu m$ .

### 3-2. Changes based on shell lengths of clam

The particle catch rates of the adult clam for the particle-size range from  $2.2$  to  $45.2 \mu m$  were more or less  $60\% \cdot h^{-1}$ , and the average size of the particles was  $3.87 \mu m$ . We further examined the influence of different shell lengths, on the particle catch-rates and their average diameters. Fig. 7 shows the catch distribution rates for clams of shell lengths 5, 10, 20, and 30 mm. The data for the 30 mm shell length shown in Fig. 7 has been adopted from Fig. 3. In case of the shell length of 5 mm, the particle catch rate was  $0.80\% \cdot h^{-1}$  corresponding to  $2.8 \mu m$  particle-size,  $0.66\% \cdot h^{-1}$  to  $11.3 \mu m$ , and  $1.1\% \cdot h^{-1}$  to  $35.9 \mu m$ , respectively. Generally the catch rates were almost uniform for particle sizes less than  $30 \mu m$ , and the average value was  $0.68\% \cdot h^{-1}$ . In case of the 10 mm shell size, the catch rate for the particle size of  $2.8 \mu m$  was  $1.51\% \cdot h^{-1}$ , that

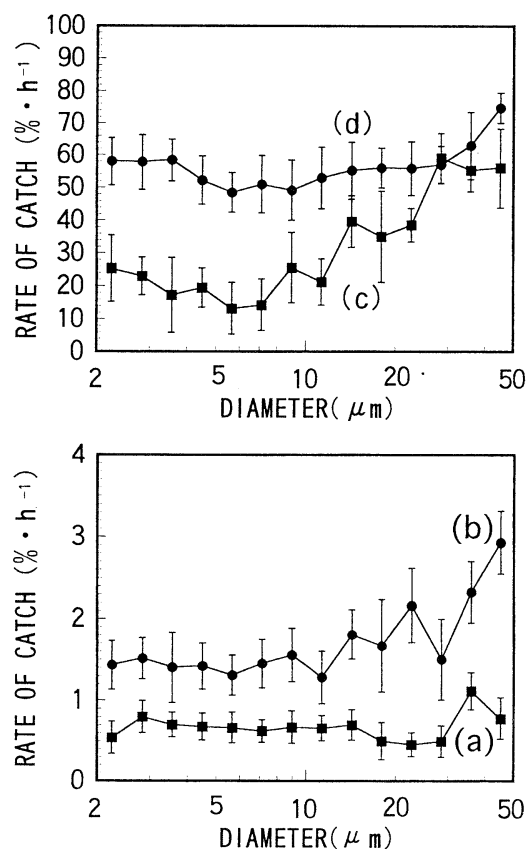


Fig. 7. Changes in the catch rates of clams dependent on their shell lengths.

The graph for the shell lengths of 5, 10, 20, and 30 mm are indicated as (a), (b), (c), and (d) in the figure. Values ( $\pm$ SD) are means of several repetitions: a=8, b=9, c=4, d=11.

for  $11.3 \mu m$  was  $1.28\% \cdot h^{-1}$ , and that for  $35.9 \mu m$  was  $2.33\% \cdot h^{-1}$ ; and in case of the 20mm, the catch rate for  $2.8 \mu m$  was  $24\% \cdot h^{-1}$ , that for  $11.3 \mu m$  was  $20\% \cdot h^{-1}$ , and that for  $35.9 \mu m$  was  $54\% \cdot h^{-1}$ . The general tendency was that the rate increased for the larger particle size. However, for the 30mm shell length group, the catch rates were again uniform, being around  $60\% \cdot h^{-1}$ , irrespective of the particle size. The average size of particles caught by clams of shell lengths 5, 10, 20, and 30 mm was 3.76, 3.80, 3.92, and  $3.87 \mu m$ , respectively. Therefore, considering the average particle sizes, it is evident that the young clam tend to catch larger sizes of suspended particles in accordance with their growth.

#### 4. Discussion

MUKAI (1993) investigated the relationship between the volume of water filtered by Manila clam and their shell length when they were either performing active filtration or not. He reported that the clam in a state of motion could filter more water as the shell length increased, while the volume of water filtered in a state of rest did not vary with shell length and the amount was only a tenth of that in a state of motion. Though the present observations are based on clam in a state of motion, we have also made observations when they were in a state of rest, and found that the filtered volumes varied remarkably with time and particle size.

Most reports till date on the intake of particles by bivalves have based the filtration rate calculations on the equation provided by JØRGENSEN (1966) and COUGHLAN (1969) which is as indicated below :

$$r = \frac{M}{n \cdot t} Ln \frac{C_0}{C_t} \quad (3)$$

where,  $r$  is the filtration rate,  $M$  is the suspension volume,  $n$  is the number of samples,  $t$  is the time lapsed,  $C_0$ ,  $C_t$  are the initial and final particle concentrations.

In our study, the particle catch rates are expressed as a percentage reduced from the initial quantity, since the range of particle size measured is broad and the sedimentation may influence the actual availability of the particles.

AKIYAMA (1985), and AOYAMA and SUZUKI (1977) investigated the filtration rate of Manila clam. They reported that the rate indicated the value of 33.8 and 33.5  $\ell \cdot \text{gN}^{-1} \cdot \text{h}^{-1}$ , respectively. In this study, we calculated the filtration rate of adult clam by equation (3) mentioned above. The result based of the total volume of particle caught by Adult is 37.0  $\ell \cdot \text{gN}^{-1} \cdot \text{h}^{-1}$ . This value is similar to afore-mentioned two results. However, we found out that the filtration rate had a great variation on quality, quantity, and size of particle. It can not be compared with both values easily.

The particle catch rates of adult clam (shell-length : 30 mm) were more or less 60%  $\cdot \text{h}^{-1}$

corresponding to the particle size range of 2.0 to 25  $\mu\text{m}$ ; above which, it tended to increase slightly.

KUSUNOKI (1965) studied the ingestion mechanism of pearl oyster, using the carbon particles of diameter 2.5 to 30  $\mu\text{m}$ , and reported that the ingestion rates were greater for the smaller particles. This is quite different from the present observations on Manila clam.

In a related study on North-East American bivalves, RIISSGÅRD (1988) reported that when the particle sizes less than 4  $\mu\text{m}$  were considered, the larger among them had higher catch-rates, whereas no such difference in catch rate was noted for the size group 5 to 10  $\mu\text{m}$ . An earlier study by JØRGENSEN and GOLDBERG (1953) on the filtration activity of *Crassostrea virginica* found that particle-sizes of 1 to 2  $\mu\text{m}$  were hardly captured, but those from 2 to 3  $\mu\text{m}$  was filtered aplenty. Thus in comparison to *Crassostrea virginica*, the clam seem to seize the smaller suspended particles ( $< 2 \mu\text{m}$ ).

In the present study we also noted the changes in catch rates with time, higher values being recorded after 60 minutes, in comparison to a third of that value when they were exposed for 180 minutes. But the catch rates for larger particles increased. MUKAI (1993) also made time based observations on the active filtration performances and found continuous alternations between a state of motion and that of rest, at different intervals from about some minutes to several hours. In our experiments, we visually inspected the samples to judge if they were in a state of rest, and if found so, the values were discarded for making necessary corrections in the catch rate calculation. It is therefore assumed that the filtration activity continued through the experiments, and the clam catch intentionally the larger suspended particles with the lapse of time.

In a study on the relation between the concentration of suspended particles and the catch rate, KUSUNOKI (1977c) disclosed that oysters mainly captured suspended particles which were more than 3  $\mu\text{m}$  in dia. when there was an abundance of the particles, but the capture size dropped to 2  $\mu\text{m}$  when there was only fewer number of suspended particles. This is similar to the observations on Manila clam.

CHIBA and OSHIMA (1957) reported that the Manila clam caught both organic and inorganic the suspended particles irrespective of their quality, provided the particle-size ranged from 3 to 4  $\mu\text{m}$ . In the current investigation employing both pellet and phytoplankton, there were no differences in catch rates corresponding to particle sizes less than 5  $\mu\text{m}$ , though it increased remarkably above that size. Judging from these results, it is considered that the Manila clam tend to catch indiscriminately the particles less than 5  $\mu\text{m}$  in size, and above that size they exhibit some preference. A study probing the relation between the captured particle size and the shell length, by FURUKAWA (1961), revealed that hard clam with the shell-length of 39.7 to 48.8 mm had a higher ability to catch suspended particles than those of the group 50.0 to 65.2 mm. However, the Manila clam could catch intentionally the larger suspended particles in accordance with their growth.

In the present study, when the Manila clam with the shell lengths of 5 to 20mm were exposed to a particle concentration of  $3 \times 10^5$  pellets/ml, the larger particles were found to be captured by clams of greater shell-lengths. This size preference by the clam noted in this study could either be due the changes in the gill characteristics or the experimental particle concentration. This has to be examined further.

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