A STUDY ON THE RATE OF FILTRATION OF THE CLAM MERETRIX CASTA (CHEMNITZ)*

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THE amount of water that passes over the gills of lamellibranchs is of considerable interest in the study of nutritional, respiratory or excretory activities of these animals. It is also of value in studying the effects of water pollution on commercially important shellfish.

The rate of water propulsion in several lamellibranchs has been studied by many a worker using direct or indirect methods. In the direct method, the water pumped is separated and measured (Galtsoff, 1926, 28; Nelson, 1935, 36; Loosanoff and Engle, 1947 and Collier, Ray, Magnitzky and Bell, 1953). The indirect method, involving the removal of particles by the filtering mechanism has been widely adopted. Calcium carbonate, colloidal graphite and unicellular algae have been used as particles in this method (Fox, Sverdrup and Cunningham, 1937; J^rgensen, 1943, 49; Rao, 1953 and Ballantine and Morton, 1956). Cole and Hepper (1954) employed neutral red solution while J<jbrensen and Goldberg (1953), Rice (1953), Chipman and Hopkins (1954) and Rice and Smith (1958) made use of radioactive phosphorus with colonial diatoms.

The present investigation was taken up with a view to studying the filtering activity of the edible clam *Meretrix casta* in different sizes and also in different salinities. This was also of interest because of the fact that the marine fish farms where these clams grow abundantly, have peculiar environmental conditions (Tampi, 1959, 60), and the study of the behaviour of these clams under such conditions is of value for clam culture in these farms.

MATERIAL AND METHODS

The clam *M. casta* was collected from the experimental marine fish farms of the Central Marine Fisheries Research Institute, Mandapam. They were brought to the laboratory and kept in sea water aquaria so as to clear them of the faeces.

Method for the determination of water filtration was essentially that of Cole and Hepper (1954) with certain modifications. Clams were kept individually in 200 or 300 ml. of fresh sea-water adjusted to the experimental salinity. When they opened and began active filtration, the water in the finger bowls was slowly changed by the neutral red solution prepared in the water of the experimental salinity. This was done by siphoning out the water from the finger bowls, and simultaneous letting in of the neutral red solution from the aspirator jar. Double the quantity of neutral red was passed through the finger bowls to ensure complete replacement of the plain sea-water. Volume of the neutral red solution in the finger

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bowls was maintained at 200 or 300 ml. as the case may be. Readings were taken at intervals of about 20 minutes and every time 10 ml. solution was pipetted out and compared with the standard in the Hellige Duboscq visual colorimeter, after the addition of a few drops of 5 % hydrochloric acid. The standard was passed into the second cup with a graduated 10 ml. pipette till both the colours matched. This modification made the whole technique simple and more accurate.

Neutral red solution employed was of the strength of 0.00025 %. For clams of the largest size 0.00075 % solution was used. Water of low salinity was prepared by the addition of the fresh water and that of high salinity by dissolving varying quantities of unpurified common salt collected from the lagoons adjoining the fish farm.

Studies were carried out on the clams belonging to the size-groups 2.2-2.4, 2.5-2.7, 3.0-3.4, 4.3-4.5 cm., and in the salinities of 8.00, 15.00, 25.00, 34.00, 45.00, 56.00 and $64.00^{\circ}/_{oo}$. In each experiment, about 25 clams were used. The data of the clams that filtered less than 70 % of the solution were not taken up for final analysis, as this would give an incorrect picture of the experimental results. For the experiments in the salinity variation, clams were acclimated to the experimental salinity, before employing them for the experiments.

RESULTS AND DISCUSSION

The rate of filtration of neutral red. There was found to be a wide variation in the filtration rate of any individual clam. The data when plotted showed a wide scatter, however, in a definite pattern. The calculated values for each size group and salinity have been shown in the graphs (Figs. 1 and 2), so as to make the data more presentable and also to give an idea about the rate of filtration. Except for the salinities of 8.00, 56.00 and 64.00 %₀, the straight line was fitted for the data of each experiment, by the method of least squares. For the salinities of 8.00 and 56.00 %₀, higher degree curve was found necessary. No curve could be fitted for the data at the salinity of 64.00 %₀ and hence the raw data are plotted as such (Fig. 3).

Following are the equations for each size group and salinity.

Size	Salinity %,,	Equation
2.2-2.4	34.00	Y=40.7656+0.2927x
2.5-2.7	? ?	Y=54.8898+0.2187x
3.0-3.4))	Y = 49.8143 + 0.299x
4.3-4.5	? ?	Y = 53.4275 + 0.3622x
2.5-2.7	8.00	$Y = -13.2550 + 1.0654x - 0.0030x^2$
	15.00	Y=40.3185+0.2218x
	25.00	Y=32.22+0.3772x
	34.00	Y=54.8898+0.2187x
	45.00	Y-41.69+0.2651x
	56.00	$Y = 28.3335 + 0.7168x - 0.0023x^2$

It may be added here that a straight line is a good fit up to about 80.00 % of the neutral red filtration. Beyond this range the observed values tend to be lower than the calculated values. These low observed values may, perhaps, be due to the retar-

elation in the capacity of the clam to retain more and more neutral red in the gills. In case of the salinities 8.00 and 56.00 $\%_0$, the filtration rate slows down considerably

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Fio. 1. The rate of filtration in different size-groups of *M. casta*. (Numbers at the end of the graphs indicate the size-groups).

at a rather early stage and the values are more or less constant beyond 60 to 80% filtration. Thus, the curve in these cases flattens after the initial rise. In case of the salinity of 64.00 %₀, the filtration is very erratic and in majority of the clams it practically stops after the initial filtration, though clams may remain open with their siphons well extended.

The rate of filtration of water. Several authors have calculated the amount of water passed through the gills from the amount of suspended material filtered by the animals. Fox *et al.* (1937) calculated this in the case of *Mytilus californianus* 5

subjected to the suspension of calcium carbonate. J^rgensen (1943, 49) derived a formula to calculate the rate of water filtration in case of *Mytilus edulis* kept in a



Fio. 2. The rate of filtratioiT of M. casta in different salinities. (Numbers at the end of the graphs indicate salinities).

suspension of algal culture and colloidal graphite. In the present investigation, the amount of water filtered was calculated from the method given below.

The values for the individual clam in each experiment were plotted and a free hand curve was drawn. Amount of neutral red filtered at the end of 1st, 2nd and



FIG. 3. The rate of filtration of *M. casta* in the salinity of 64.00 % •

3rd hour was read from these graphs. The amount of water filtered per minute by each clam was obtained by substituting the above values of neutral red in a formula derived as follows.

The derivation of the formula is based on the following assumptions.

- (1) The rate of filtration of the clam remains constant during the hour under consideration.
- (2) The neutral red in the solution is completely absorbed when it passes through the clam.

(3) Twenty ml. of solution, removed for colorimetric comparison during the hour, is assumed to have been removed at the end of the hour and hence is taken into account while proceeding with the calculation of rate for the following hour.

Now, if' x' ml. is the quantity of solution to start with and ' p' is the quantity of neutral red per ml. of solution at the beginning of experiment, the total quantity of neutral red in solution is 'xp' ml. If ' a ' ml. is the rate of filtration per minute during the first hour, the quantity of neutral red absorbed during the first minute is ' ap ' ml. Remaining quantity of neutral red is ' p(x-a)' ml. and the quantity per ml. is ' p(x-a)/x.' The consumption during the second minute on the above lines is ' ap (x—a)/x' ml. and the remaining quantity will be' p(x-a)(x-a)/x' ml., the quantity of neutral red per ml. at the end of the first hour will be $(x-a)^{60}/x^{fo}$.' At the beginning of the second hour 20 ml. of the solution would have been removed leaving (x—20) ml as the total quantity of solution. The quantity of neutral red is ' (x—20) $p(x-a)^{60}/x^{fi}$ ml. If ' b ' ml. is the rate of filtration of the clam during the second hour, it can be shown, similarly, that the quantity of neutral red per ml. at the end of the second hour is ' P { (x-a)⁸⁰/x⁶⁰ } { (x-20-b)⁶⁰/(x-20)¹⁰ } '. This formula can also be extended to the third hour in order to cover the total period of the experiment.

In this connection, it may be stated that *Voxet al.* (1937) and J^rgensen (1943,49) have assumed that as the water passes through the gills of lamellibranchs, all the suspended material therein would be completely removed. They calculated the rate of filtration of water on this assumption. Rice *et al.* (1958) expressed that the rate of filtration of water calculated by the methods of the former workers, would be different from the actual pumping rate.

The formula derived here using neutral red, is apparently more accurate than by using colloidal graphite, calcium carbonate or algal suspension; as the neutral red forms a homogeneous solution unlike the latter substances. Cole and Hepper (1954), the originators of the technique also claim that the amount of neutral red removed from the water is directly dependent upon the volume of water pumped through the gills. This is independent of whether or not mucus is secreted by the animal as against the case with colloidal graphite.

The amount of water filtered per minute during 1st, 2nd and 3rd hours, in the clams of different size-groups and in different salinities have been shown in Tables 1 and 2. It would be seen that the rate of water propulsion is maximum during the first hour and gradually decreases during the subsequent hours. However, the difference between the rate during second and third hour is not much. Similar observations have been made by J^rgensen (1949) in case of *Mytilus edulis*, kept in a suspension of colloidal graphite, flagellates and *Nitzschia*. This decrease in the rate, he attributes to the heterogeneity in the particle size in graphite and also to the change in the pore size of the gills so that, the gills which act as filters may be more or less permeable to particles of the same size. He also suspects overloading of gills with micro-organisms which retard the rate of filtration. J^rgensen (1960) admits that the rate of filtration of water will differ with the efficiency of particle retention in normal and undisturbed lamellibranchs as against disturbed ones. He further suggests that the high and varying porosity recorded by several workers in case of mussels and oysters is typical of disturbed animals. Thus, undisturbed filter feeding lamellibranchs will efficiently retain particles even of a few microns in size.

means that perhaps, the rates of flow measured in the, laboratory by using any suspended material, may be different from that in natural condition.

TABLE 1

Mean rate of water filtration per minute for first, second and third hours in different sizegroups of M. casta

No.	Size cm.	Salinity °/00	No. of Clams	Mean rate per minute ml.	Variance
		FIRST HO	UR		
1 2 3 4	2.2-2.4 2.5-2.7 3.0-3.4 4.3-4.5	34.00 ≫	23 22 22 23	3.13 4.32 4.36 7.93	0.28 1.69 1.57 2.94
		SECOND HO	UR		
1 2 3 4	2.2-2.4 2.5-2.7 3.0-3.4 4.3-4.5	34.00 »»	23 14 18 16	2.15 2.62 3.18 6.93	0.43 0.68 0.37 10.73
		THIRD HOU	JR		
1 2 3 4	2.2-2.4 2.5-2.7 3.0-3.4 4.3-4.5	34.00 »»	22 8 15 ——-E	1.38 2.50 2.64 xperiment was over— 1	0.29 0.92 0.76

It would be further evident from Table 1 that the rate of filtration is directly related to the size of the clam. Larger the clam, more is the rate of filtration per minute. Fox *et al.* (1937) and Chipman and Hopkins (1954) made similar observations on *Mytilus calif ornianus* and scallops. However, the latter authors found lower rate of filtration per gram of tissue in case of larger scallops than smaller ones. Works of Rice and Smith (1958) support these observations.

In the clam *M. casta*, the total amount of water filtered per hour is less than either in scallops or mussels of approximately the same size. Fox *et al.* (1937) noted, in case of *M. calif ornianus*, the average of 6.4 l/hour of water for the size 178 mm. and the average of 14 l/hour for the size 60 to 82 mm. Willemsen (1952) observed an average filtration rate of 1.81 l/hour for *M. edulis* of 80 mm. length and a rate of 0.5 l/hour in case of *Cardium edule* of 30 to 40 mm. in length. The bay scallops of the size 38-44 mm. filters an average of 3.26, l/hour of water andjjjose of the size 47-48 filters 8.21 l/hour (Chipman and Hopkins, 1954). In *M. meretrix*, the clams of the size 2.2-2.4 cm. filter about 0.188 l/hour of water while those of the size 4.-3 4.5 cm. filter average about 0.476 l/hour. The maximum amount of water (0.786, l/hour) was filtered by the clam of the size 4.5 cm. which is lower than that observed by the above workers in scallops and *Mytilus*. However, it may be mentioned here, that reasons for the variations in the rates of filtration in different lamellibranchs, observed by these workers, may perhaps be found in different experimental techniques employed by these workers (J^rgensen, 1960).

It is well known that the rate of filtration has relation with the feeding activity of the clam. The amount of food available to suspension feeding lamellibranch is

TABLE 2

Size cm.	Salinity %0	No. of Clams	Mean rate per minute ml.	Variance
	FIRST H	OUR		
2.5-2.7 ^{↓ ↓} ≫ ≫	$\begin{array}{c} 8.00 \\ 15.00 \\ 25.00 \\ 34.00 \\ 45.00 \\ 56.00 \\ 64.00 \end{array}$	22 25 21 22 23 21 12	2.19 2.60 3.18 4.32 2.95 3.37 2.12	$\begin{array}{c} 0.15 \\ 0.39 \\ 0.42 \\ 1.69 \\ 0.29 \\ 0.63 \\ 0.46 \end{array}$
	SECOND 1	HOUR		
2.5-2.7),),),),), ,,	$\begin{array}{c} 8.00 \\ 15.00 \\ 25.00 \\ 34.00 \\ 45.00 \\ 56.00 \\ 64.00 \end{array}$	22 23 21 14 23 20 12	$ \begin{array}{r} 1.89 \\ 1.53 \\ 1.53 \\ 2.62 \\ 1.69 \\ 2.06 \\ 0.75 \\ \end{array} $	$\begin{array}{c} 0.06 \\ 0.31 \\ 0.12 \\ 0.68 \\ 0.42 \\ 0.30 \\ 0.31 \end{array}$
	THIRD H	OUR		
2.5-2.7 js i* *j t* j*	$\begin{array}{c} 8.00 \\ 15.00 \\ 25.00 \\ 34.00 \\ 45.00 \\ 56.00 \\ 64.00 \end{array}$	22 23 17 8 22 18 12	$ \begin{array}{r} 1.07\\ 1.26\\ 1.38\\ 2.50\\ 1.68\\ 1.28\\ 0.63 \end{array} $	$\begin{array}{c} 0.08\\ 0.29\\ 0.29\\ 0.92\\ 1.00\\ 0.42\\ 0.24 \end{array}$
	Size cm. 2.5-2.7 ¹¹ ² 2.5-2.7 ¹⁾ ³ ³ ³ ³ ³ ³ ³ ³	Size cm. Salinity % o FIRST H 2.5-2.7 8.00 11 25.00 34.00 a 34.00 34.00 b 64.00 56.00 b 56.00 56.00 b 56.00 56.00 b 64.00 56.00 b 34.00 56.00 b 56.00 56.00 b 64.00 56.00	Size cm. Salinity %0 No. of Clams FIRST HOUR 2.5-2.7 8.00 22 11 15.00 25 25.00 21 34.00 2 45.00 23 3 56.00 21 3 64.00 12 SECOND HOUR Colspan="2">THIRD HOUR 2.5-2.7 8.00 22 3 56.00 21 3 25.00 21 3 34.00 14 3 56.00 20 3 64.00 12 THIRD HOUR C.5-2.7 8.00 22 3 56.00 20 3 56.00 23 3 56.00 23 3 56.00 12 THIRD HOUR 2.5-2.7 8.00 22 3 15.00 23 3	Size cm. Salinity %0 No. of Clams Mean rate per minute ml. 2.5-2.7 8.00 22 2.19 1 25.00 21 3.18 * 34.00 22 4.32 * 34.00 21 3.18 * 34.00 21 3.37 * 56.00 21 3.37 * 64.00 12 2.12 SECOND HOUR 23 1.53 * 34.00 14 2.62 1* 25.00 21 1.53 * 34.00 14 2.62 1* 45.00 23 1.53 * 34.00 14 2.62 1* 64.00 12 0.75 * 56.00 23 1.69 1* 64.00 12 0.75 * 45.00 23 1.69 1* 56.00 12 0.75 * 34.00

Mean rate of water filtration per minute, for first, second and third hours in different salinities in M. casta

determined by the rate at which the surrounding water is transported through the gills, (.tyrgensen, 1960). From the foregoing account, it appears that *M. casta* from the marine fish farm has low rate of feeding. In view of this, it is interesting to study the index of condition and chemical analyses of this clam, as both are dependent upon the feeding activity.

Effects of varying salinities on the filtration rates of lamellibranchs have been studied by Dodgson (1928), Cole and Hepper (1954) and Nagabhushanam (1956). These workers observed a depressing effect of low salinities on the rate of filtration. In the present investigation, it was found that the rate of filtration is adversely affected both at low and high salinities (Table 2). In the salinity of 8.00 %₀ the rate is 2.19 ml. per minute which increases with the increasing salinity and is highest (4.32 ml/minute) at the salinity of 34.00 %₀. The rate remains fairly high even at the salinities of 45.00 and 56.00 %₀. At the salinity of 64.00 %₀, the filtration rate retards and becomes very erratic. At this salinity, the clams were seen fully extending their

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siphons without filtration even for considerable time. Clams would withdraw their siphons slowly even when disturbed, as against their usual habit of quickly closing the valves even with slight disturbance. This suggests that, very high salinities may have benumbing effect on the clams.

The foregoing observations indicate that in *M. casta* there is a considerable degree of physiological adaptation to the changing salinities and the rate of filtration is not much affected in the salinity range of 25.00 to $56.00 \circ/_{00}$. In this connection, it may be mentioned that, in the marine fish farms, from where these clams have been brought, the salinity goes very high $(50.0 \circ/_{00})$ during summer and remains above $40.0 \circ/_{00}$ f^{or} 5 months in a year. It rarely falls below $30.0 \circ/_{00}$. It is likely that the clams here are habituated to wide variations in the salinities. Cole and Hepper (1954) have observed that mussels may become habituated to conditions of salinity to which they are more frequently and regularly exposed.

Rate of acclimation of M. casta *to different salinities.* This was studied by keeping 25 clams belonging to the size group 2.5-2.7 cm. in one litre water of the experimental salinity. In each experiment 100 clams were used. The experiment was continued till at least 20.0% clams opened. However, in case of the salinity $64.0 \circ /_{00}$ the observations were discontinued when 10.0% of the clams filtered. This last day was taken as the day of acclimation. The water in the troughs was changed daily. Readings were noted only for the first three hours every day, as it was found that the clams begin to close after a few hours. Table 3 gives the rate of acclimation of these clams to different experimental salinities. It could be seen from the table that the clams opened on the first day in the salinities 15.00 to $56.00 \circ /_{00}$ while in the salinities of 8.00 and $64.00 \circ /_{00}$ they could not acclimate easily. The percentage of clams opened on the first day is also fairly high in all the above salinities, except that of 15.00 and $56.00 \circ /_{00}$. These results further support the observations that *M. casta* has a greater degree of physiological adaptation in the salinity range 25.00 to $56.00 \circ /_{00}$.

TABLE 3

No.	Salinity °/00	Average per- centage of clams opened for the first three hours of the first day	Percentage of clams opened for the first three hours on the last day	The day clams opened first and their per- centage	Mortality during experi- ment
1 2 3 4 5 6 7	$\begin{array}{c} 8.00\\ 15.00\\ 25.00\\ 34.00\\ 45.00\\ 56.00\\ 64.00\end{array}$	nil 28.00 64.67 79.34 91.00 1.34 nil	23.34 C17» 53.67 (5)* 55.34 (5)» 75.34 (5)* 65.34 (5)* 49.56 (9)* 10.00 (22)*	3 (2.5%) 1 (28.0%) 1 (64.67%) 1 79.34%) 1 (91.00%) 1 (1.34%) 4 (3.00%)	5 1 nil 1 nil 15

The acclimation of M. casta to the different experimental salinities. {All values are the averages of only first three hours}

•Number of days the experiment was run,

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SUMMARY

The rate of filtration in *Meretrix casta* is studied by adopting Cole and Hepper's neutral red technique. The studies indicate that the rate of filtration increases with the size. However, it is much less than that observed in other lamellibranchs of the same size. The rate of filtration is rapid during first hour of the experiment and gradually falls during the subsequent hours. The rate falls in excessive low and high salinities, while the clams can readily adapt themselves for a wide range of salinities, from 25.00 to $56.00^{\circ}/_{00}$. It appears that very high concentration of salts in the water have more or less benumbing effect on the clams. The experiments on the acclimation of clams to different experimental salinities support these observations.

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