Proc. Indian Acad. Sci. (Anim. Sci.), Vol. 98, No. 1, January 1989, pp. 65–76.

Byssal threads of *Mytilopsis sallei* (Recluz) and their adhesive strength

M UDHAYAKUMAR and A A KARANDE
Biology Division, Naval Chemical and Metallurgical Laboratory, Tiger Gate, Bombay 400 023, India

MS received 28 September 1988; revised 12 December 1988

Abstract. The dreissenid mussel *Mytilopsis sallei* (Recluz) settles on man made structures in confined harbour waters. The mussel unlike some mytilids is never found to have settled on coastal sea bed. It shows a good capacity to produce a byssus apparatus and develops a new one every time it settles afresh. Younger mussels develop byssus apparatus at shorter intervals and therefore move more often. Adults are relatively passive. Byssal thread development is influenced by the environmental factors and also by the quality of surface. The mussel achieves better adhesion on polar surfaces like slate and glass than on teflon. Tensile strength of adhesive threads is very poor as compared to other mytilids. This possibly is one of the reasons for its absence on sea beds where sea water turbulence is very high.

Keywords. Marine; biofouling; mytilids; byssal adhesion.

1. Introduction

Mussel *Mytilopsis sallei* has almost established itself in Indian tropical waters. This mussel is not found in open channel waters of Bombay and seems to prefer man-made structures in confined water of harbours. If this mussel can settle or not on the near shore sea bed is yet unknown. In wet basins a fouling debris weighing as much as 10 kg/m^2 is generated as a consequence of heavy growth of *M. sallei* (figure 1).

M. sallei has excellent capacity to produce byssus threads and as many as 1000 threads are counted from a single byssus apparatus of an adult mussel pulled off from a submerged surface. The mussel in search of a desired surface can voluntarily shed its byssal apparatus (Udhayakumar and Karande 1986) and develop a new one every time it settles afresh.

Gross morphology of the byssal apparatus of this mussel conforms to that of most of the other mytilids (Bairati and Vitellaro-Zuccarello 1974a, b; Brown 1952). It consists of 3 portions viz. root embedded in the tissues of the foot, a stem and a number of byssus threads each ending in an adhesive disc (figure 2). It is presumed that the adhesive discs in this mussel are amongst the smallest in surface area (0·21 sq.mm). Karande and Menon (1975) observed that *M. sallei* can withstand extreme salinity of 2‰ as long as 4 months of continuous exposure and show seemingly normal activity up to 38°C during 24 h exposure. They have also noted that the mussel survives as long as 10 months on a single dietary source of *Dunaliella primolecta* under laboratory conditions.

Presuming that *M. sallei* is a calm water inhabitant, its reported migration from the Central Atlantic to the Indian Ocean (Morton 1981), is an interesting dispersal phenomenon.

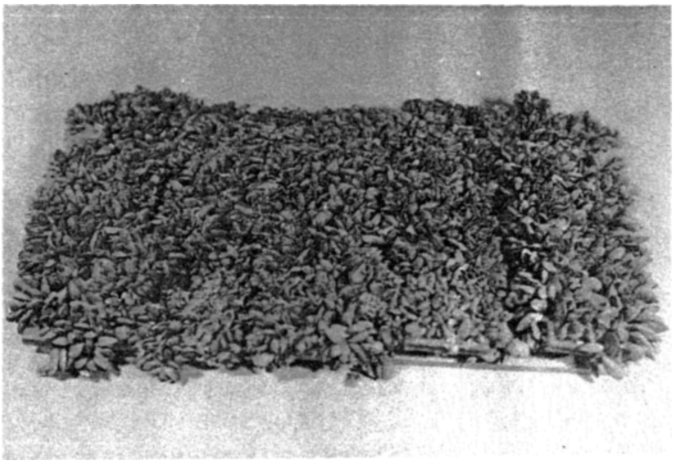

Figure 1. Heavy growth of *M. sallei* in confined polluted waters in Bombay harbour (120 days).

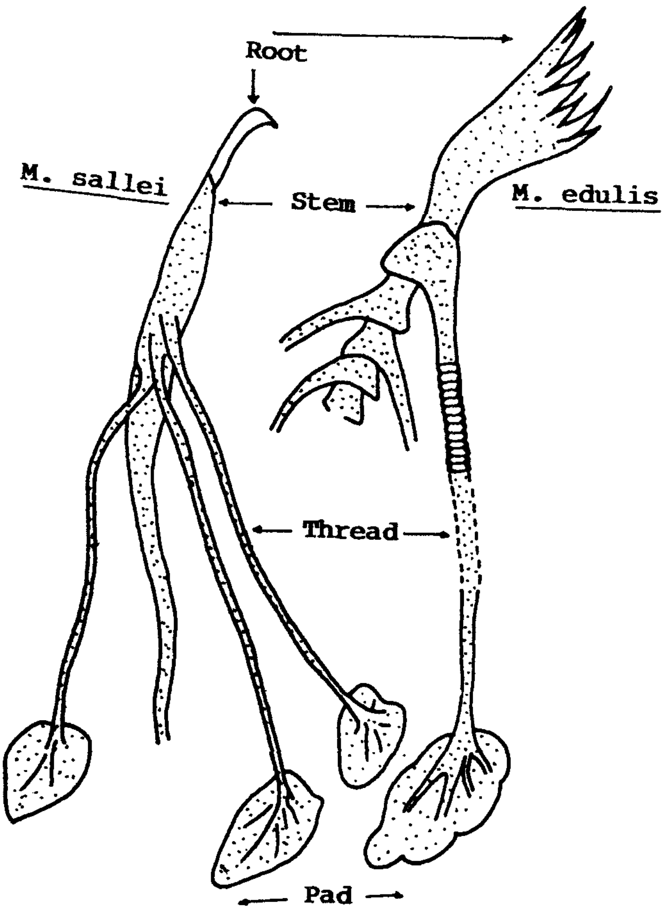

Figure 2. Byssus apparatus of *M. edulis* (after Young and Crisp 1982) and *M. sallei*.

2. Materials and methods

The mussels were collected from the water front structures for the field study and from the fouling coupons for the laboratory experiments. In the laboratory, organisms were kept in an aerated aquarium tanks at 28°C temperature and fed on algal diet of *D. primolecta.*

For counting byssus threads, the mussels were carefully dissected to separate the byssus apparatus. The numbers of threads were counted under stereomicroscope.

The behaviour of mussels on various offered surfaces was examined in 4 litre glass aquaria. Seawater aged for 48 h was used and the mussels were not fed during the experimental period.

For determining adhesive strengths of the byssus apparatus, a method recommended by Gubbay (1983) was adopted. The mussels attached on various surfaces were loaded statically on an Instron tensile testing machine. The load was applied at 20 mm/min and the maximum load borne prior to catastrophic failure resulting in complete detachment of the mussel from the substratum was recorded.

The details of various experiments performed are mentioned alongwith the observations noted under various sections.

3. Results

3.1 *Byssal thread production*

Among the sessile marine organisms, the species securing attachment by byssus threads are at advantage as compared to the others like balanids and polychaetes whose adherence to the surface is permanent. Mytilids can move from place to place if subjected to environmental stresses. It is, therefore, observed that these organisms produce varying number of byssus threads and the adhesive strengths of the byssus vary seasonally and with the nature of environments (Pieters *et al* 1978; Price 1980, 1982; Waite 1983).

3.1a *Field observations*: Table 1 summarises the data on the extent of byssus threads produced at various size classes of the mussels collected from the study

Table 1. Size dependent variations in numbers of byssal threads.

Observations		Size classes in (mm) shell length			
		10	15	20	>21
Set I	n	21	33	49	15
	x̄	141·9 ± 22	159·0 ± 16·4	329·1 ± 31·4	318·0 ± 82
Set II	n	16	52	31	12
	x̄	71·0 ± 10·9	132·4 ± 11	202·2 ± 20·3	309·5 ± 34·7
Set III	n	38	45	42	23
	x̄	104·1 ± 8·4	163·2 ± 11·7	245·0 ± 12·9	328·0 ± 25·2

n, Number of observations; x̄, average number of byssus threads formed.

station. It is seen from this table that the number of threads produced is more in the larger organisms. For instance in set I, comprising 3 independent observations, average numbers of threads counted in each size classes 1, 2, 3 and 4 have been 141, 159, 329 and 318 respectively. A similar trend is noted in sets II and III observations as well.

Field data were further analysed to ascertain if there existed any seasonal influence on the thread production. It is seen from table 2 that the number of threads produced during the monsoon was always numerically more than those produced during pre and post monsoon months. The threads produced during the monsoon were significantly more than those produced during post monsoon period.

3.1b *Laboratory observations*: Table 3 summarises the observations made on the ability of the mussel to produce byssal threads during 120 h under ambient laboratory conditions. Seven to ten individuals of each of 3 size classes, i.e. 5, 15 and 20 mm were used. It was observed that within 120 h the mussels dislodged and resettled on as many as 9 occasions, each time producing varying number of byssus threads. Average numbers of threads produced generally increased with the size class but having reached a size of more than 20 mm, ability to produce fresh byssus threads at consecutive resettlements was reduced. It was evident, therefore, that the younger mussels which displayed better ability to produce threads moved more often than the adult mussels. The latter because of their inability to produce adequate threads at shorter intervals, displayed restricted movements. The adults lack capacity to produce fresh byssal apparatus, but once having properly settled in the field conditions produce larger number of threads than the younger mussels.

3.2 *Salinity and byssus formation*

In these experiments influence of salinity on the development of byssal threads was examined. The surfaces offered for the attachment were gravel, glass and the mussel shells. Five mussels were exposed to each of 10, 20, 35 and 40‰ salinities for a period of 7 days where they often changed places of attachment.

Table 4 gives average numbers of byssus threads formed under different salinity conditions, all the other conditions remaining the same as the ambient laboratory

Table 2. Seasonal variations in the development of byssal threads.

Season	Size class (i) < 15 mm (ii) > 15 mm	'*P*' Value	Statistical analysis: Season	Statistical analysis: Remarks
Monsoon	(i) 147 ± 12	0·05	Monsoon/Post-monsoon	S
	(ii) 325 ± 27	0·02		
Post-monsoon	(i) 117 ± 9	0·10	Post-monsoon/Pre-monsoon	NS
	(ii) 238 ± 19	0·20		
Pre-monsoon	(i) 141 ± 9	0·50	Monsoon/Pre-monsoon	NS
	(ii) 274 ± 12	0·10		

S, Significant; NS, not significant.

Table 3. Byssal thread production under laboratory conditions.

Size class (mm)	Specimen	No. of byssus produced at every fresh attachment during 120 h									Cumulative no. of threads	Remarks
		1	2	3	4	5	6	7	8	9		
5	1	3	9	20	60	—	—	—	—	—	92	Av. cumulative/individual = 66 ± 12·9
	2	5	10	16	11	13	18	6	18	25	122	
	3	8	25	—	—	—	—	—	—	—	33	
	4	7	11	13	20	18	7	—	—	—	76	
	5	4	7	11	23	—	—	—	—	—	45	
	6	6	7	10	10	—	—	—	—	—	33	
	7	4	4	7	11	5	—	—	—	—	31	
	8	10	7	6	6	8	—	—	—	—	37	
	9	4	21	16	8	16	8	46	6	—	125	
15	1	9	15	15	25	41	19	6	—	—	130	Av. cumulative/individual = 92·85 ± 19·9
	2	25	42	38	12	26	12	—	—	—	155	
	3	8	6	6	—	—	—	—	—	—	20	
	4	6	18	34	14	19	29	20	9	—	149	
	5	2	12	12	9	14	22	9	—	—	80	
	6	36	7	—	—	—	—	—	—	—	43	
	7	21	20	32	—	—	—	—	—	—	73	
20	1	31	—	—	—	—	—	—	—	—	31	Av. cumulative/individual = 18·14 ± 4·36
	2	24	13	—	—	—	—	—	—	—	37	
	3	1	6	11	—	—	—	—	—	—	18	
	4	10	—	—	—	—	—	—	—	—	10	
	5	13	—	—	—	—	—	—	—	—	13	
	6	12	—	—	—	—	—	—	—	—	12	
	7	6	—	—	—	—	—	—	—	—	6	

Table 4. Byssus threads produced under various salinity conditions.

	Salinity (‰)			
	10	20	35	40
No. of threads (Cumulative of 5 mussels)	404 ± 28	330 ± 45	278 ± 24	229 ± 53
Influence of salinity	10/35 salinity $P < 0.005$ S	20/35 salinity $P < 0.50$ NS		35/40 salinity $P < 0.50$ NS

S, Significant; NS, not significant.

environment. Assuming 35‰ as a prevailing normal sea water salinity, an increase in number of threads was noted in organisms exposed to lowest salinity of 10‰. There were no significant variations in numbers at salinities 20 and 40‰.

3.3 *Temperature and byssus formation*

The mussels were subjected to 4 different temperatures of 15, 20, 30 and 35°C. It was observed (table 5) that there was no significant influence of raised temperature on the thread formation. At lower temperatures of 15 and 20°C, however, the numbers of threads formed were significantly small than those formed at ambient temperature of 30°C.

3.4 *Substrate quality and attachment*

3.4a *Mussel behaviour on independent surfaces*: The preference of the mussel for 2 polar surfaces namely slate and glass and a non-polar teflon was examined. The mussels also opted to settle on the shells of the fellow experimental organisms and therefore the settlement on this surface was also considered in these experiments. Each experiment was carried out for a period of 3 weeks continuously.

Three groups of mussels identified as (a), (b) and (c), each containing 15 mussels were placed on each of the 3 surfaces by rotation for a period of one week. For instance (see table 6), group (a) mussels were placed on slate in the first week, then on glass in the second week and on teflon in the third week. In each group there had been a steady fall in thread making activity during each week. It is for this reason that as recommended by Young (1983) 3 groups had to be used and placed in turn on each surface. Table 6 shows that on slate, glass and teflon more threads were formed than on mussel shell. Table 6 (row 7) gives ratios of numbers of discs formed on offered surfaces to number of discs on shells. All the 3 surfaces, slate, glass and teflon, in that order were preferred to shell surface.

3.4b *Choice of surface for byssal attachment*: Three pairs of surfaces like slate/glass, wax/slate and glass/wax were offered to examine mussels choice for the surface. Eight mussels were placed on each surface and the experiment was repeated for 5 weeks, each week mussels being subjected to different choice between pairs of surfaces.

Table 5. Byssus threads produced under various temperature conditions.

	Temperature (°C)			
	15	20	30	35
No. of threads (cumulative of 5 mussels)	$7·6 \pm 1·8$	$46·8 \pm 9·7$	$208·1 \pm 21·7$	$171·7 \pm 25·6$
Influence of temperature	15/20°C $P < 0·0005$ HS	20/30°C $P < 0·00005$ HS		30/35°C $P < 0·05$ NS

HS, Highly significant; NS, not significant.

Table 7 shows that in the 5 weekly trials, the numbers of discs formed on wax were always smaller than those on glass and slate. It is also observed that the numbers of pads per unit area of surface were always greater by several order of magnitude on the polar surfaces of slate and glass.

3.4c *Surface texture and byssal deposition*: To assess the influence of surface texture on the deposition of byssal threads, 7 surfaces were offered for the settlement. Three of these were roughened and offered with their smooth counterparts. Table 8 shows that glass, fibreglass and perspex when roughened, induced higher thread formation as compared to their smooth surfaces.

3.4d *Byssal deposition on gravel*: An experimental set up for carrying out this experiment is illustrated in figure 3. It is seen from table 9 that the mussels not only preferred gravel surface to teflon but in doing so produced more adhesive discs on gravel. Some mussels adhered partly to gravel and partly to teflon but even in this event more discs were adhered to the former.

3.4e *Surface quality and disc area*: Twenty mussels of varying shell sizes were allowed to settle on glass surface. Figure 4A shows a relationship between mean area of discs formed by each individual and its shell length. The disc area was found to grow linearly with the increasing shell size.

Ten mussels each were allowed to grow on polar and non-polar surfaces for a period of 10 days. Average areas of discs of the mussels settled on slate, glass, perspex and teflon were 0·13, 0·21, 0·26 and 0·16 mm^2. No correlation therefore was noted between disc area and the polarity of the surface.

3.5 *Adhesive strength of byssal attachment*

In order to measure adhesive strength of the mussel, a method earlier adopted by Gubbay (1983) was followed. Figure 4B, C shows the relationship between the areas covered by discs on different surfaces and the forces required for the dislodgement of the individual mussels. There appears to be no relation between the two. It is inferred therefore that though the disc area increases with the growth of the shell, an adhesive bond secured by individual mussel is not proportional to the area of adhesion achieved. This implies that the discs lose adhesive bond as they grow old, necessitating continuous production of fresh threads by this mussel.

Table 6. Number of newly formed threads and location of attachments of pads on different surfaces.

Surface	Slate			Glass			P.T.F.E.			Total no. of threads
No. of pads	On slate I	On mussel II	Total III	On glass IV	On mussel V	Total VI	On P.T.F.E. VII	On mussel VIII	Total IX	On all surfaces (% on mussels) X
Week 1	(a) 582	92	674	(b) 232	0	232	(c) 219	148	367	1273 (18·85)
2	(c) 394	102	496	(a) 221	120	341	(b) 439	107	546	1383 (23·78)
3	(b) 136	14	150	(c) 257	27	284	(a) 118	23	141	575 (11·13)
Total	1112	208		710	147		776	278		
Ratio of no. of pads on surface to nos. on mussels	5·34:1			4·82:1			2·79:1			

Table 7. Numbers of pads formed on 3 pairs of surfaces slate/glass, wax/slate and glass/wax during 5 successive weeks.

Week	Pairs of surface	No. of pads	Pads/cm^2
1	Slate and glass	7 and 186	0·02 and 0·96
	Wax and slate	27 and 276	0·11 and 1·40
	Glass and wax	182 and 1	0·94 and 0·004
2	Slate and glass	130 and 36	0·53 and 0·18
	Wax and slate	7 and 73	0·02 and 0·37
	Glass and wax	24 and 1	0·12 and 0·004
3	Slate and glass	92 and 82	0·38 and 0·42
	Wax and slate	9 and 76	0·03 and 0·39
	Glass and wax	291 and 1	1·5 and 0·004
4	Slate and glass	169 and 62	0·69 and 0·3
	Wax and slate	5 and 313	0·02 and 1·29
	Glass and wax	174 and 4	0·9 and 0·01
5	Slate and glass	7 and 30	0·02 and 0·15
	Wax and slate	1 and 167	0·004 and 0·86
	Glass and wax	138 and 17	0·76 and 0·07

Table 8. Byssal threads produced during 7 days under laboratory condition.

	No. of threads Surface condition	
Surface	Smooth	Rough
Slate	32·5 ± 6	—
Asbestos	31·0 ± 6·42	—
Balasa wood	15·0 ± 2·89	—
Glass	12·0 ± 2·41	32·0 ± 11·9
Fibre glass	12·0 ± 4·14	31·0 ± 9·59
Perspex	27·17 ± 4·17	48·42 ± 7·42
Wax	8·0 ± 1	—

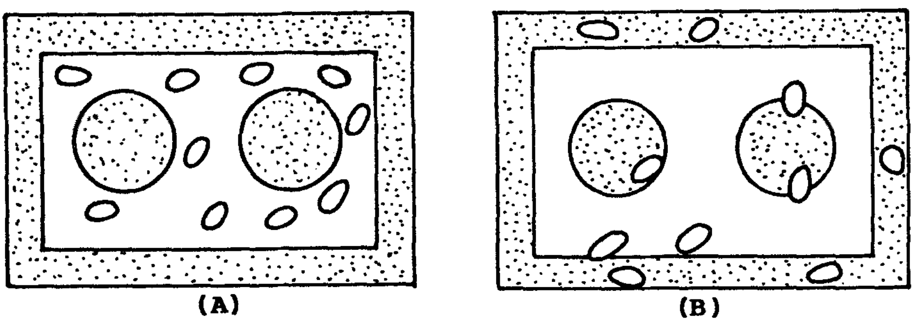

Figure 3. Schematic representation of the experimental set up for ascertaining the choice of *M. sallei* for teflon (total area 1394 sq.cm) and gravel (total area 766 sq.cm) substrates. **A.** Position at the beginning of experiment. **B.** Position at the end of experiment.

Figure 4B, C also reveals that the forces required for dislodging the mussels from the two polar surfaces namely slate and glass are greater (up to 112 g) than that

Table 9. Choice of *M. sallei* between teflon and gravel surfaces.

Experiment	Gravel: No. of mussel attached	Gravel: Av. no. of thread/ mussel	Teflon: No. of mussel attached	Teflon: Av. no. of thread/ mussel	Preference to gravel: As per mussels attached	Preference to gravel: As per threads formed
1	15	37·8	3	26·6		
2	10	25·7	4	16·5	$t = 2{\cdot}79$ $P < 0{\cdot}05$ S	$t = 2{\cdot}861$ $P < 0{\cdot}05$ S
3	11	28·3	5	13·0		
Average	12 ± 1·5	30·6 ± 3·6	6·3 ± 1·3	18·7 ± 4·0		

S, Significant.

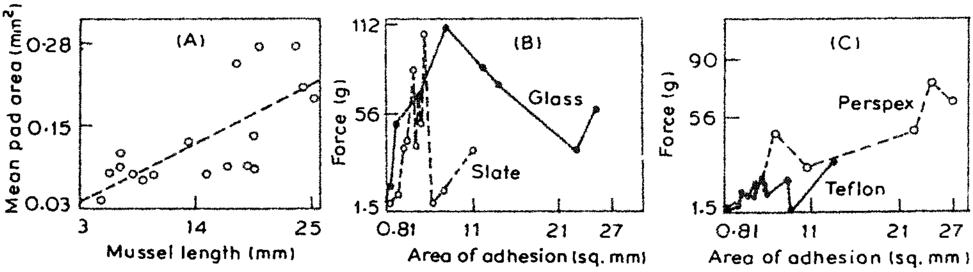

Figure 4. Relationship between (A) *M. sallei* shell length and mean adhesive pad area, (B) area of adhesion and the force required for dislodging the mussel from slate and glass surfaces and (C) area of adhesion and the force required for dislodging the mussel from perspex and teflon (PTFE) surfaces.

needed for teflon (32 g). Udhayakumar and Karande (1986) have reported that the force required for the removal of *M. sallei* is far less ($0{\cdot}09 \times 10^8$ Nm^{-2}) than that required for big mussel *M. edulis*. In the latter species a force of $3{\cdot}6 \times 10^8$ Nm^{-2} is required to dislodge one single byssal thread (Young and Crisp 1982). The force required to pull off the whole animal therefore is expected to be very high.

4. Discussion

M. sallei on a given surface readily forms byssus within 7 min, the successive threads emerging at intervals of 2 or 3 min. In this respect its ability to secure first attachment is as good as *M. edulis* (Young 1983). The mussel can voluntarily shed its byssus apparatus as often as 6 times within 24 h and 9 times in 120 h. This is an advantageous situation so far as the freedom of the mussel to move and resettle is concerned.

M. sallei produces a larger number of adhesive threads during the monsoon period. It was noted that more threads were formed at lower salinities of 10‰ than at normal saline water. An ability of the mussel to sustain low and widely varying salinity has, as a matter of fact, helped this species to establish itself in Indian waters (Karande and Menon 1975). This mussel produced less number of byssal threads at lower temperatures of 15 and 20°C as compared to normal ambient temperature of 30°C. Waite (1985) has, however, observed that 'none of these (earlier) studies conclusively demonstrates that byssal formation is specifically influenced by environmental factors, instead, the changes in byssal formation are often used to measure changes in the byssal rate of metabolism'.

No chemo-reception mechanism is studied in these mussels, however, a tuft of cillia on foot in *M. edulis* is described as being mechanoreceptors (Young 1983). Seed (1967) concluded that smooth surfaces such as tufnol attracted very few larvae, the maximum settlement being on roughened or fibrous surfaces. Both Maas Geesteranus (1942) and Chipperfield (1953) made similar observations. Young and Crisp (1982) inferred that 'as far as possible mussels avoid attaching the byssus to low energy surfaces, and do so only when high energy surfaces are not made available'.

In *M. sallei* it was noted that rough surface did induce higher development of byssal threads. It was observed that the thread formation on non-polar wax surface was significantly poor as compared to slate or glass surface. In *M. sallei*, the shell was found to be a least preferred substrate as was a teflon surface.

Earlier Udhayakumar and Karande (1986) had observed that in *M. sallei* there was no correlation between the disc areas and polarities of the offered surfaces. They, however, observed that the forces required to pull off the mussel from glass and slate were greater (up to 112 g) than that needed for teflon (32 g).

M. sallei are found to have ability to settle on gravel but this by itself is not adequate to maintain a sustained adhesion to the sea-bed. Besides smaller adhesive discs (0·21 mm^2), a smaller body size and a shorter foot, one more factor that possibly limits the distribution of *M. sallei* in open waters is its inability to sustain a relatively high turbulence earlier reported by Swami and Karande (1988). The tensile strength of byssal complex of *M. sallei* as a whole is around $0{\cdot}09 \times 10^8$ Nm^{-2} (Udhayakumar and Karande 1986). On the other hand the strength of one single byssus thread of *M. edulis* is $3{\cdot}6 \times 10^8$ Nm^{-2}. Despite this superior tensile strength of the byssal thread of *M. edulis*, Price (1982) observes that in that species 'wave action emerges as a major determinant of byssal attachment strength'. This could as well be true in case of *M. sallei*.

Acknowledgement

Authors are thankful to Dr R Krishnan, for his helpful support.

References

Bairati A and Vitellaro-Zuccarello L 1974a Ultrastructure of the byssal apparatus of *Mytilus galloprovincialis* III. Analysis of the byssal characteristics by polarized light microscopy; *J. Submicrosc. Cytol.* **6** 367–379

Bairati A and Vitellaro-Zuccarello L 1974b Ultrastructure of the byssal apparatus of *Mytilus galloprovincialis* II. Observations by micro-dissections and scanning electron microscopy; *Mar. Biol.* **28** 145–158

Brown C H 1952 Some structural protein of *Mytilus edulis*; *Q. J. Microsc. Sci.* **93** 487–502

Chipperfield P N J 1953 Observations on the breeding and settlement of *Mytilus edulis* L. in British waters; *J. Mar. Biol. Assoc. UK* **32** 449–476

Gubbay S 1983 Compressive and adhesive strengths of a variety of British barnacles; *J. Mar. Biol. Assoc. UK* **63** 541–555

Karande A A and Menon K B 1975 *Mytilopsis sallei* a fresh immigrant in Indian harbours; *Bull. Dept. Mar. Sci. Univ. Cochin* 7 455–466

Maas Geesteranus R A 1942 On the formation of banks by *Mytilus edulis*; *Arch. Neerl. Zool.* **6** 283–326

Morton B 1981 The biology and functional morphology of *Mytilopsis sallei* (Recluz) (Bivalvia: Drissenacea) fouling Visakhapatnam harbour; *J. Moll. Stud.* **47** 25–42

Pieters H, Kluytmans J H, Zurburg W and Zandee D I 1978 The influence of seasonal changes on energy metabolism in *Mytilus edulis* (L.) I. Growth rate and biochemical composition in relation to environmental parameters and spawning; in *Cyclic phenomena in marine plants and animals*: *Proc. 13th Europ. Mar. Biol. Symp., Isle of Man* (eds) E Naylor and R G Hartnoll (Pergamon Press) pp 285–292

Price H A 1980 Seasonal variation in the strength of byssal attachments of the common mussel *Mytilus edulis* L.; *J. Mar. Biol. Assoc. UK* **60** 1035–1037

Price H A 1982 An analysis of factors determining seasonal variation in the byssal strength of *Mytilus edulis*; *J. Mar. Biol. Assoc. UK* **62** 147–155

Seed R 1967 *Ecology of Mytilus edulis (L.) on exposed shores*, Ph.D. thesis, University of Leeds, UK

Swami B S and Karande A A 1988 Recruitment and growth of biofouling invertebrates during monsoon in Bombay water, west coast of India; *Indian J. Mar. Sci.* **17** 143–149

Udhayakumar M and Karande A A 1986 Adhesive strengths of some biofouling organisms; *Curr. Sci.* **55** 656–658

Waite J H 1983 Adhesion in byssally attached bivalves; *Biol. Rev.* **58** 209–231

Waite J H 1985 Catechol oxidase in the byssus of the common mussel *Mytilus edulis* L.; *J. Mar. Biol. Assoc. UK* **65** 359–371

Young G A and Crisp D J 1982 Marine animals and adhesion; *Adhesion* **6** 19–39

Young G A 1983 Response to and selection between, firm substrata by *Mytilus edulis*; *J. Mar. Biol. Assoc. UK* **63** 653–659