



Seasonal and tidal changes in the length of the crystalline style intertidally living *Macoma balthica* (Mollusca, Bivalvia) *

H. Hummel¹, W. de Bruin², G. Nieuwland² and I. F. Hummel-Poel¹

24283

¹ Delta Institute for Hydrobiological Research, Vierstraat 28, 4401 EA Yerseke, The Netherlands

² Netherlands Institute for Sea Research, P.O. Box 59, 1790 AB Den Burg, Texel, The Netherlands

Abstract

Samples of *Macoma balthica* were collected during 1978 and 1979 from the westernmost part of the Dutch Wadden Sea. The length of the crystalline style in the stomach of this intertidal bivalve was studied through tidal and annual cycles. The length of the style was ca. half the shell length and changed slightly with the season. Maximum lengths were recorded in spring, the season of high food intake, and minimal values in autumn and winter. Changes in the length of the style also exhibited a tidal cycle, but with two maxima and two minima. Tidal fluctuations in style length amounted to ca. 5% only. The influence of enzymes and waste products from the digestive diverticula, the food intake as well as the influence of the pH on the length of the style are discussed.

the stomach at the end of the emersion period. The style reforms after the feeding period, at the end of the submersion period or during the emersion.

Surprisingly, the changes in the length of the crystalline style have never been studied simultaneously with food intake or stomach content. The period of food intake was mostly estimated indirectly, e.g. from the adductor muscle activity.

This paper reports on changes, according to season and tide, in the length of the crystalline style of *Macoma balthica* living on a tidal flat in the Dutch Wadden Sea. Simultaneously determined stomach contents and rates of food intake are described by Hummel (1985a).

Introduction

The crystalline style is a long hyaline rod in the stomach of almost all bivalves. Several authors have attached a central role to it in the abrasion of food particles and in the secretion of extracellular enzymes (Yonge 1949, Purchon 1968, 1971, Morton 1973). As a consequence of rotation, the style mixes and triturates the food (Nelson 1918, Yonge 1926, 1949, Allen 1958, Purchon 1968, Morton 1970b); however, this rotation has often been disputed over the last decade (Purchon 1971, Kristensen 1972a, Bernhard 1973, Hughes 1977). The style is nevertheless important because it contains a variety of enzymes (Kristensen 1972b, Morton 1983) which are released by partial or complete dissolution of the style. Changes in the length, by dissolution and reformation, have often been observed in intertidal molluscs, and are related to the tidal rhythms of food intake and digestion (see review in Morton 1973). Dissolution is initiated by the release of waste products from the digestive diverticula into

Materials and methods

Samples of *Macoma balthica* were taken during 1978 and 1979 at Balgzand, a tidal flat area in the westernmost part of the Dutch Wadden Sea. The sampling station was situated 1½ km from the coast and at –30 cm MTL (mean tidal level). On the average, the station was covered by the tide for 7½ h and exposed for 5 h. The aerial mean tidal range is between –77 cm and +57 cm MTL.

Samples 12.5 to 20 mm long were collected at intervals of 1 to 2 h. *Macoma balthica* live buried in the sediment at a depth of 1 to 10 cm (Hulscher 1973, Chambers and Milne 1975, Reading and McGrorty 1978). Sediment samples were obtained during the period of submersion by means of a manual corer or a small adapted cockle-scraper, and during emersion by digging. These sediment samples were sieved and the bivalve individuals were taken quickly on board the research vessel. Four stomachs were dissected immediately under magnification. The total length of the crystalline style, including the anterior tip (see Kristensen 1972a, Dean 1980 for the morphology of the style), was measured immediately after dissection to the nearest 0.1 mm. The shell length was also measured to the nearest 0.1 mm.

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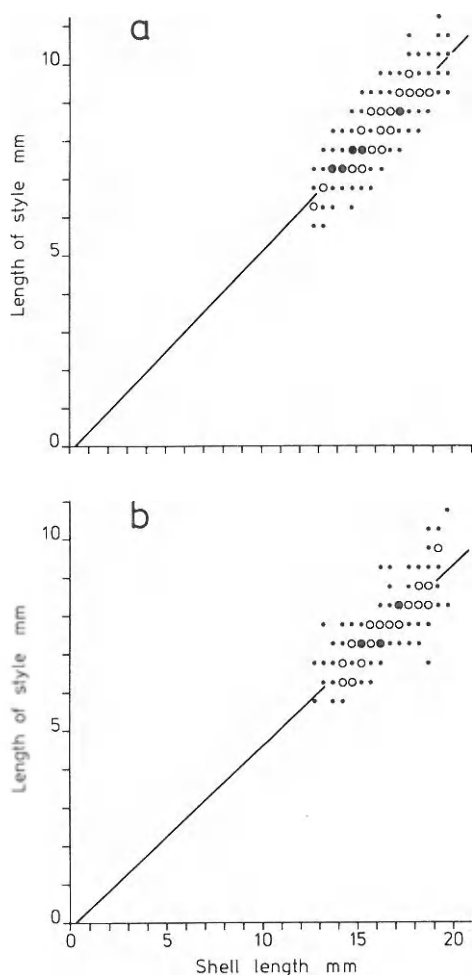


Fig. 1. *Macoma balthica*. Relation between length of crystalline style and length of shell during spring (a) April, May and June and autumn and winter (b) September to March. Full line represents the best fitting regression through all individual observations (•: 1 to 5 observations; ○: 6 to 10 observations; ●: 11 to 15 observations)

Concomitant amounts of chlorophyll *a* in the stomach and rates of food intake were estimated, as described by Hummel (1985a).

Results

Style length versus shell length

The style length of *Macoma balthica* showed a linear relationship to the shell length (Fig. 1). This was so during both the growing season (Fig. 1a) and the rest of the year (Fig. 1b). The intercept of $y(a)$ of the best fitting lines did not differ significantly from 0 ($p > 0.10$), and, therefore the style length can be expressed as a percentage of the shell length (relative style length).

Linear relationships between the style length and shell length have also been observed in the related species *Abra nitida* and *Abra alba* (Kristensen 1972a) with a style length about one third the shell length. Moreover, a linear relation-

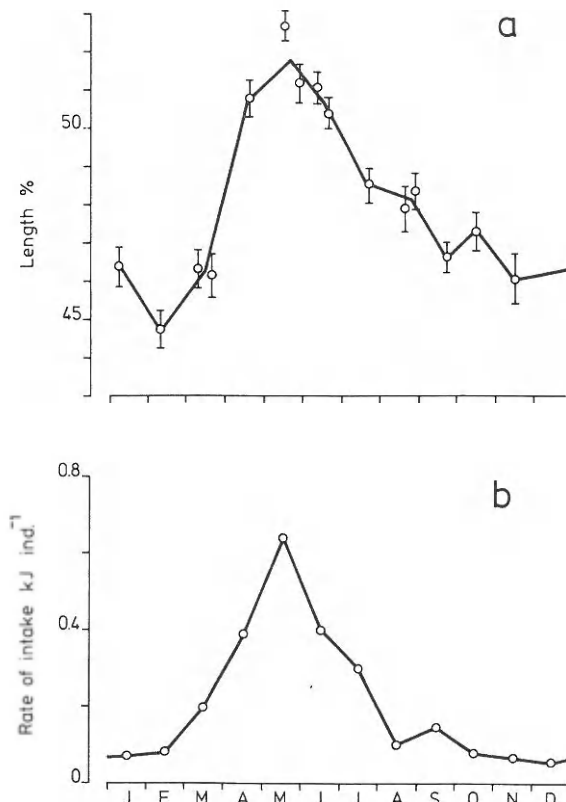


Fig. 2. *Macoma balthica*. (a) Relative length of the style and (b) rate of intake of chlorophyll *a* (in kJ per individual per month; after Hummel 1985b) through the year. Indicated are daily (a) or monthly (b) mean values together with the standard error (a). Full line connects mean values of all observations in a month

ship was found between the dry weight of the style and the total wet weight of *Crassostrea gigas* (Bernard 1973).

Seasonal variations

The style length of *Macoma balthica* varied with the season (Fig. 2a). The relative length was highest (above 50% of the shell length) during spring, coinciding with the season of high food intake (Fig. 2b, Hummel 1985a) and somatic growth (Beukema and de Bruin 1977). Minimal values for relative style length were observed during autumn and winter (below 47%), coinciding with the season of low rates of food intake (Fig. 2b, Hummel 1985a), zero length growth and loss of weight of soft parts (Beukema and de Bruin 1977).

The difference in style length between spring (April, May and June) and the "off-season" was highly significant (Student's *t*-test; $p \leq 0.001$). This seasonal difference amounted to 5–8% of the shell length or to 10–16% of the style length.

Tidal and daily variations

The relative length of the style changed with a consistent cyclical pattern during a tidal cycle (Fig. 3). A significant

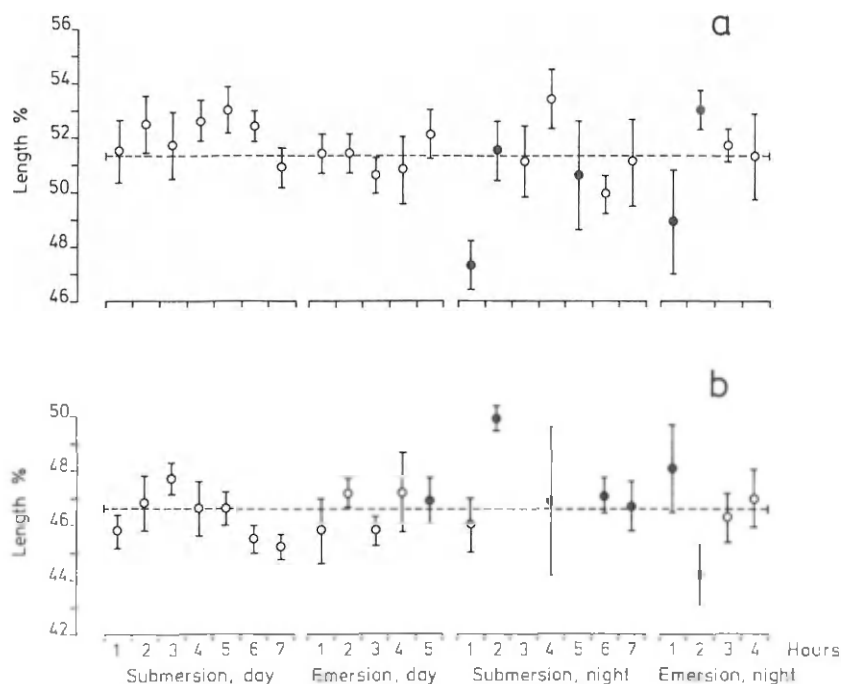


Fig. 3. *Macoma balthica*. Fluctuations in relative length of crystalline style during submersion and emersion periods in day and night separately shown for (a) spring (April, May, June) and (b) autumn and winter (September to March). Indicated are mean values for hourly intervals together with standard error. Open symbols indicate means of four or more observations, filled symbols of less than four observations

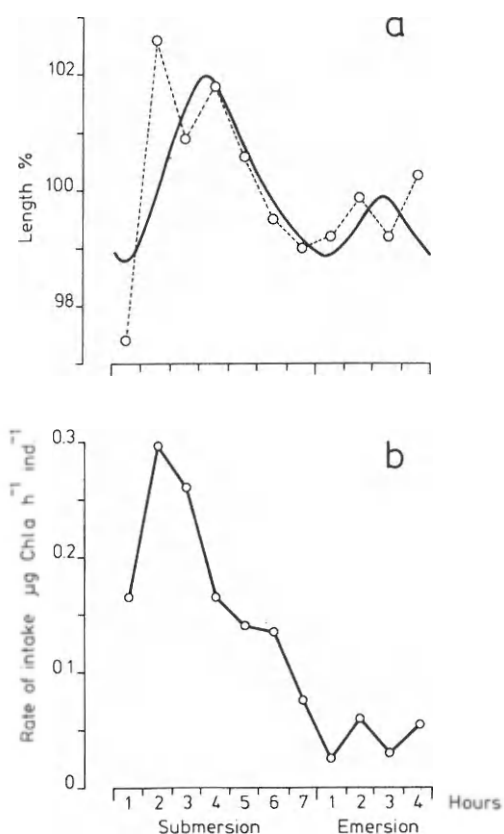


Fig. 4. *Macoma balthica*. Fluctuations in (a) length of style during submersion and emersion (as a percentage of daily mean length) and (b) rate of intake of chlorophyll *a* (in μg per individual per hour) (b after Hummel 1985 a). Full line in (a) represents Fourier description of data

($p < 0.01$) Fourier description of the changes in the style length showed two maxima and two minima during one tidal cycle. This pattern was similar during the growing season (Fig. 3a) and the rest of the year (Fig. 3b) and also for both day and night. Therefore the data were indexed and pooled (Fig. 4a). The most dominant cycle in the Fourier analysis (Fig. 4a) had a period equal to one tidal cycle.

At the beginning of the submersion period the style was short, then reached a maximum in the middle of this period and decreased again to low values at the end. During the subsequent emersion period the style lengthened slowly, only to decrease again rapidly when the tide came in.

The maximal difference in the average style length during a tidal cycle (Fig. 4a) amounted to 5% of the style length. The maximal hourly rate of style reformation, observed during the first hours of submersion, was about $5\% \text{ h}^{-1}$ of the total style length (Fig. 4a).

Discussion

The changes in style length of *Macoma balthica* have to be explained as changes in the balance between style formation and dissolution. The rate of change in style length varied from -3 to $+5\%$ per hour. Thus the maximal net formation rate would be 5% per hour (but probably more). Accepting that this rate is equal to the gross formation rate and is also constant, the style would have a turn-over time of 20 h. This turn-over time does not differ much from that found for *M. balthica* by Kristensen (1972a) and is between that for other species (Table 1).

Records on seasonal differences in style length in molluscs appear to be scant. Kristensen (1972a) observed in *Abra* that the styles are present throughout the year, but during winter these styles were somewhat soft.

Tidal rhythms in changes of the style length have been described for several molluscs (Table 2). In all but one mollusc species studied, a tidal rhythm in the changes of the style length was found, with only one maximum and one minimum. The style lengthened at the beginning of the submersion or feeding period and attained a maximal length during or at the end of that period. Reduction of the style occurred at the end of the submersion period and during emersion or when the bivalve stopped feeding. This results in a minimal length during emersion or at the very beginning of submersion.

This general trend is partly in contrast with the model proposed by Morton (1973). On the basis of a literature review, Morton put forward a theory in which tidally changing processes of food intake, digestion and style dissolution were connected. Similar to the general trend, he described also a reduction of the style length at the end of the emersion period. However, he stated that the style would not be reformed during the feeding period, but only when the individ-

ual stops feeding. Thus a maximum style length would then be reached during emersion. It is surprising that Morton did not incorporate reformation of the style during submersion or the feeding period in his model, since he referred to similar studies as shown in our Table 2.

The changes in style length we found for *Macoma balthica* resemble those of the general trend during submersion and those of Morton's model during emersion. This means that two maxima and two minima are found during one tidal cycle. The minimum at the beginning and the maximum close to the end of the emersion period are, however, inconspicuous and, therefore, such a trend might not have been found in the earlier studies mentioned in Table 2.

The style is formed in the style sac (Morton 1970b, Kristensen 1972a, Dean 1980) and is secreted in coarse and fine lamellae. Although data on changes in the rate of style formation are not available, an average rate of style formation can be calculated indirectly. In the style of *Macoma balthica* at least 40 finer lamellae are visible, whereas a turn-over time for the whole style of 24 h is mentioned (Table 1; Kristensen 1972a). Thus, the new style material is probably secreted in 30 min periods or less. These periods are too short to explain the observed changes in style length.

On the other hand, style dissolution has been studied much more intensively than style formation. For style dissolution three underlying and interrelated processes are described: (a) the arrival of acid digestive enzymes and waste products from the digestive diverticula into the stomach, (b) the acidity of the stomach content and (c) the feeding activity (Table 2). For *Macoma balthica* the first process is described by Morton (1970a). Spherules containing enzymes and digestive waste products are formed during emersion in the digestive diverticula. They are expelled into the stomach at the end of emersion. Extracts from the digestive diverticula dissolve the style (Kristensen 1972a). Thus, the minimal style length at the end of emersion or beginning of

Table 1. *Macoma balthica*. Turn-over-time of the style

Species	Turn-over time (h)	Author
<i>Macoma balthica</i>	24	Kristensen 1972a
<i>Abra nitida</i>	4	Kristensen 1972a
<i>Crassostrea gigas</i>	3	Bernard 1973
<i>Choromytilus meridionalis</i>	24	Seiderer et al. 1982
<i>Perna perna</i>	136	Seiderer et al. 1982
<i>Crassostrea virginica</i>	≤ 12	Lucas and Newell 1984

Table 2. *Macoma balthica*. Rhythmicity and causes of changes in the length of the style of molluscs

Author	Species	Position ¹	Rhythm ²	Formation ³	Max length	Dissolution	Min length	Cause of changes ⁴
Morton 1956	<i>Lasaea rubra</i>	i	t	s (beg)	s	s (end)	e (end)	food, d.d.
Morton 1970b	<i>Cardium edule</i>	i	t	s		e		food, pH, d.d.
Kristensen 1972a	<i>Macoma balthica</i>							food, pH, d.d.
Langton and Gabbott 1974	<i>Ostrea edulis</i>	i	t (3)		s (end)		s (beg)	food, pH
Langton 1977	<i>Mytilus edulis</i>	i	t (1-2)	s	s (end)	e	s (beg)	pH
McHenry et al. 1983	<i>Mytilus edulis</i>	i	t		s (end)			
Lucas and Newell 1984	<i>Crassostrea virginica</i>	i	t		s		c (-)	
Robertsen 1979	<i>Ilyanassa obsoleta</i> (gastropoda)	i	t		s		e (-)	food
Morton 1971	<i>Ostrea edulis</i>	s	t				feed (beg)	food, pH, d.d.
Mathers 1976	<i>Pecten maximus</i>	s	t					
Robinson and Langton 1980	<i>Mercenaria mercenaria</i>	s	n					

¹ i: intertidal; s: subtidal

² Rhythm of changes in style length: t: tidal; n: no rhythm found; (x): rhythm is x hours out of phase with tidal cycle

³ s: submersion; e: emersion; (beg): beginning of; (end): end of; feed: during feeding period, (-) style absent;

⁴ food: presence or absence of food; d.d.: presence of fragmentation spherules from the digestive diverticula; pH: acidity

submersion is most probably caused by dissolution from products out of the digestive diverticula. This phenomenon is obviously so strong and universal among marine molluscs that it is found in almost all studies described (Table 2).

Secondly, the dissolution time of the style is influenced by the pH (Table 3). At higher pH the dissolution time decreased. At the pH of seawater, i.e., around 8, dissolution time is lowest. At this pH the style may dissolve in 1 to 3 h. Normally, the pH of the style is around 6 to 7 (Table 4). Changes in the pH showed a tidal cycle (Table 4). These changes resemble the changes in style length; a minimum around the end of emersion, a maximum halfway during the submersion period. The minimum coincides with the release of acid waste products from the digestive diverticula. The maximum coincides with the intake of food. The intake of food is the third factor influencing the dissolution of the style although its influence is only indirect. With the intake of food, seawater with a relatively high pH is taken in too, and at the same time acid waste products and enzymes from the digestive diverticula are diluted. For *Macoma balthica* the rate of food intake is described by Hummel (1985a, Fig. 4b).

Table 3. *Macoma balthica*. Dissolution time (hours) of the style in relation to pH

Species	pH			Author
	3.5–4	6	7.8–8.5	
<i>Macoma balthica</i>	96		1	Kristensen 1972 a
<i>Ostrea edulis</i>	2.8	1.5	0.9	Mathers 1974
<i>Pecten maximus</i>		4.6	3.0	Mathers 1976
<i>Venerupis decussata</i>	4.8	3.8	1.0	Mathers et al. 1979
<i>Chlamys varia</i>	4.2	0.7	0.3	Mathers et al. 1979

The succession of events influencing the length of the style in *Macoma balthica* might then tentatively be as follows: At the end of the emersion period enzymes and acid waste products are expelled from the digestive diverticula into the stomach. The enzymes increase the dissolution of the style strongly, and the style reaches its minimal length. Yet, because of the acid waste products, the pH around the style drops simultaneously. When this happens the enzymes are inactive and the dissolution rate drops drastically while the length of the style increases. Then, due to a strong food intake (Fig. 4b), the pH, and thus the dissolution rate, increase. Halfway the submersion the dissolution is again stronger than the style formation, and the length of the style starts to decrease again. In the second half of the submersion period the food intake decreases and remains low during emersion. Consequently the pH decreases to normal levels, and with it the dissolution rate. This leads to a small net increase of the style length. At the end of the emersion a new cycle starts.

Similarly, the seasonal changes in style length might be influenced indirectly by the rate of food intake. Outside the growing season the food intake of *Macoma balthica* is low (Fig. 2b). A lower food intake is most probably accompanied by a lower production of acid waste products in the digestive diverticula. Therefore, the pH around the style remains (on an average) higher, and with it the dissolution of the style. Moreover, a lower food intake may result in less energy directed towards the formation of new style material. Together, the stronger dissolution and the lower formation rates may cause a smaller style outside the growing season.

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Table 4. *Macoma balthica*. pH of style or style-sac and in stomach of bivalves

Author	Species	Position ¹	pH style ²		pH stomach ²	
			minimum	maximum	minimum	maximum
Morton 1970 b	<i>Cardium edule</i>	i			6.2–6.9	e. (a) s. (b)
Purchon 1971	<i>Scrobicularia plana</i>	i	6.5			
	<i>Macoma balthica</i>	i	6.7			
	<i>Cardium edule</i>	i	6.4			
Mathers 1974	<i>Ostrea edulis</i>	i	6.1–6.9	s (beg)	s (half), (b)	
Langton and	<i>Ostrea edulis</i>	i	6.3–7.2 (6.6)		5.5–7.2 (6.0)	
Gabbot 1974	<i>Crassostrea angulata</i>	i	6.3–6.9 (6.6)		5.9–6.9 (6.5)	
Langton 1977	<i>Mytilus edulis</i>	i	6.2–6.7	e (end) (half) + s (beg), (a)		
Morton 1971	<i>Ostrea edulis</i>	s	6.4–7.2 (7.0)	feed (beg), (a)		
Mathers 1976	<i>Pecten maximus</i>	s	5.5–6.7 (6.2)	(b)	5.2–6.0 (5.6)	
Mathers et al.	<i>Venerupis decussata</i>	s	5.8–7.1 (6.4)		5.4–6.9 (6.1)	
1979	<i>Clamys varia</i>	s	6.2–7.1 (6.4)	around high water	5.7–6.3 (6.0)	around high water

¹ i: intertidal; s: subtidal

² (a): minimum caused by products from the digestive diverticula; (b): maximum caused by the arrival of fresh food and seawater; s: submersion; e: emersion; feed: feeding period; (beg) = beginning of x, x (half) = halfway x, x (end) = at the end of x

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