Elsevier

JEM 00783

The effects of extended periods of drainage and submersion on condition and mortality of benthic animals*

H. Hummel[1], A. Meijboom[2] and L. de Wolf[1]

[1]*Delta Institute for Hydrobiological Research, Vierstraat 28, 4401EA Yerseke, The Netherlands;* [2]*Research Institute for Nature Management, Texel, The Netherlands*

(Received 15 April 1986; revision received 22 August 1986; accepted 25 August 1986)

Abstract: Temporary closure of the storm-surge barrier in the Oosterschelde estuary (The Netherlands) affects the tidal amplitude and rates of tidal currents. The aim of this study was to assess experimentally the effects of tidal manipulation on the numbers and condition of intertidal benthic animals. To this end undisturbed sediment cores and isolated animals were exposed to prolonged drained conditions (ebb) or submerged in stagnant water. Part of the drained sediment cores received a daily extra supply of simulated rain water (rained cores).

Permanent submersion did not affect the benthic animals. Most species suffered heavily from drainage, irrespective of an extra supply of tap water. The smaller animals without shells, such as anemones and small polychaetes, were the most susceptible to drainage, gastropods the least. The mortality rate was highest during the summer, somewhat lower in spring and lowest in autumn and winter. The decrease in water content and the change in salinity in the sediment, as observed in the drained and rained sediment cores, did not contribute to the survival (or mortality rate) of the animals. At drainage the ambient air-temperature and the glycogen content of the animals determined their mortality rate. High temperatures (25 to 30 °C) and a low glycogen content increased the mortality rate. No decrease in the glycogen content of the animals during the stress-periods was observed.

Key words: Drainage; Submersion; Benthos; Mortality; Tidal manangement; Glycogen

Introduction

To protect the area around the Dutch Oosterschelde estuary from floods a storm-surge barrier will be completed in 1987. Temporary closure of this storm-surge barrier will affect the tidal amplitude and rates of tidal currents in the estuary. As a consequence, benthic animals in the estuary may experience, depending on their position in the tidal zone, extended periods of dry (ebb) conditions (drainage) or submersion in possibly stagnant water.

The aim of this study was to assess the mortality and changes in condition of the benthic animals caused by artificial changes in the tidal rhythm. To this end large undisturbed cores of sediment, including the animals, were sampled and transported to an outdoor sea-water tank with an artificial tidal cycle. Moreover, animals were taken

* Contribution No. 357 from Delta Institute for Hydrobiological Research.

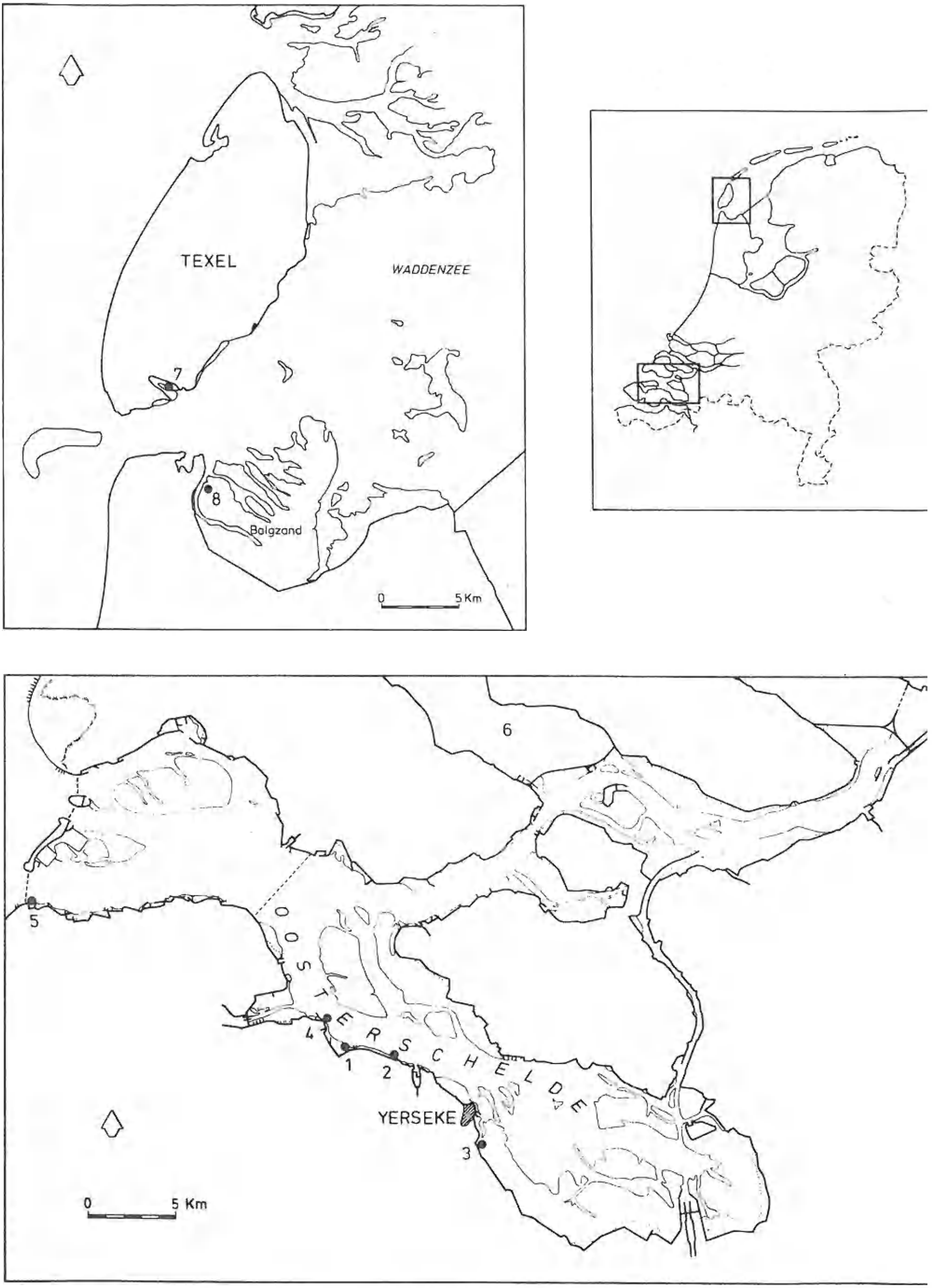

Fig. 1. Location of the sampling stations: the animals sampled in the southwestern part of the Netherlands were studied in Yerseke; the cores collected in the Wadden Sea were studied at Texel.

from their natural habitat and placed in sea-water aquaria in the laboratory. The changes in numbers and condition of the drained and submerged animals were followed.

Materials and Methods

The experiments were partly carried out in aquaria at Yerseke, and partly in a large outdoor tank at Texel (Fig. 1). The animals (studied in Yerseke) and the sediment cores (studied in Texel) were collected from tidal flats, except for *Ostrea edulis* which was collected in the Grevelingen, a stagnant brackish lake (Station 6, Fig. 1).

The other animals used in the laboratory experiments were collected in the Oosterschelde estuary (Fig. 1, Table I). *Cerastoderma edule*, *Arenicola marina* and *Nephtys hombergii* were collected at Station 1, *Sagartia troglodytes* and *Diadumene cincta* at Station 2 (*Diadumene* during autumn at Station 4), *Littorina littorea* at Station 3, and *Mytilus edulis* at Station 5. The animals were kept in cylindrical aquaria 30 cm wide.

Table I

Sampling dates, temperatures and duration of the laboratory experiments at Yerseke and outdoor experiments at Texel.

Yerseke				
Season	Autumn	Winter	Spring	Summer
Start of experiment	17 Nov. 1983	18 Jan. 1984	19 Apr. 1984	11 July 1984
Duration of stress-period (days)	14	14	14	14
Duration of recovery-period	28	28	27	14
Average temperature (°C)	9	3	10	19, 22
Texel				
Season	Summer	Autumn	Winter	Spring
Start of experiment	4 July 1983	31 Oct. 1983	12 Jan. 1984	17 Apr. 1984
Duration of stress-period (days)	14	14	14	14
Duration of recovery-period	28	28	27	28
Average temperature (°C)	20	5	5	10

Aquaria with polychaetes were filled with a 15-cm thick layer of sediment, those with *Cerastoderma edule* with a layer of sediment 5 cm thick. The aquaria were placed in two temperature rooms, one with a constant, the other, to simulate the normal daily fluctuation, with a varying temperature (in the daytime 5 °C above, at night 5 °C below average). During summer both temperature rooms had a varying temperature. The temperature was controlled within a range of 2 °C. The aquaria were connected to a continuously recirculating sea-water system of 500 l. The sea water was filtered on a Filtrox 5 μm filter. After filtration a culture of the flagellate algae *Isochrysis galbana* Parke and some powdered mussel meat were added to the water, to give food concentrations of 0.5 to 2 mg POC per l.

The sediment cores used in the outdoor tank experiments at Texel were obtained from the western Waddenzee with a box corer (Fig. 1, Table I). The species composition of the benthic fauna on the tidal flats in the Waddenzee and in the Oosterschelde estuary are quite similar (Beukema, 1976; Fortuin, 1981). The sampling surface of the box corer was 0.25 m^2 and the sampling depth 30 to 50 cm. After sampling, the cores were placed on racks in the outdoor tank (Fig. 2). Racks of different heights were used to store simultaneously drained (high racks), submerged (low racks) and control cores in the tank. Sea water could be stored in an adjacent tank. The water level in the tank with sediment cores could be adjusted by pumping water from and to the "water"-tank. A change of the tidal cycle and amplitude was established by means of electronic clocks and water-level indicators connected to the pumps. The tanks contained 300 m^3 sea water. The sea water was continuously renewed. The tidal range in the western Waddenzee is small, mean high water being + 54 cm and mean low water being − 82 cm from mean tidal level. Station 7 was situated at + 20 cm and Station 8 at − 70 cm from MTL. Two stations were sampled to obtain a representative range of species.

The animals in the aquaria (at Yerseke) were divided into three groups and the sediment cores (at Texel) into four groups:

(1) a group drained for 2 wk (drained),

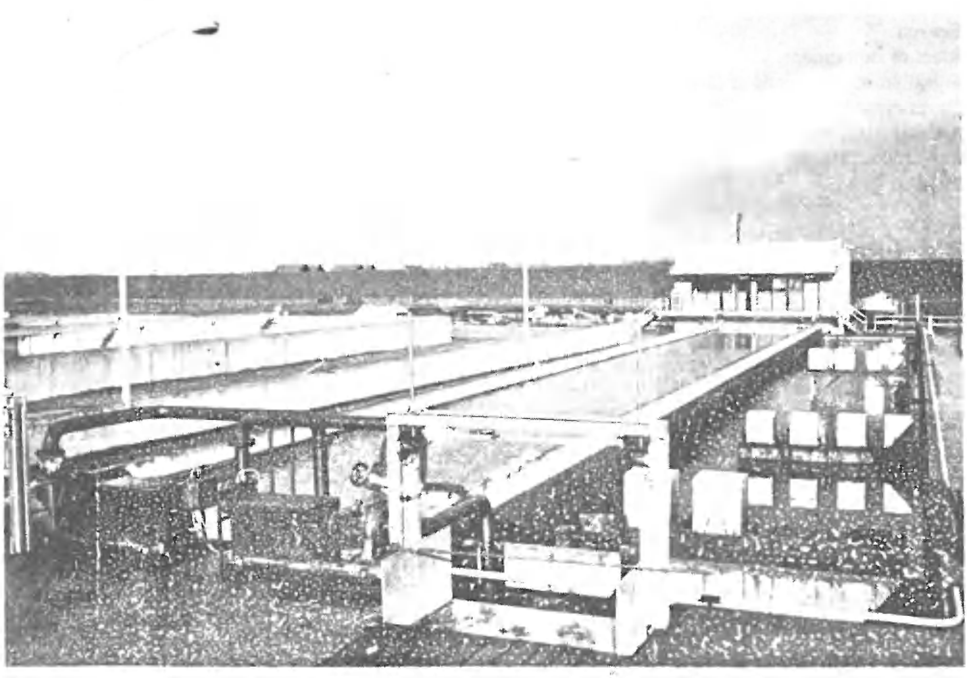

Fig. 2. Overview of the experimental set-up with sediment cores in the outdoor tanks of the Research Institute for Nature Management at Texel: from a slide of A. Meijboom.

(2) a group submerged for 2 wk in almost stagnant water (submerged),

(3) a group subjected to a normal tidal cycle (control), and

(4) a group, only for the cores, drained and to which a daily amount of tap water equivalent to 2 mm rain was added on top of the sediment (rained).

The stress-period of 2 wk was followed by a recovery period of 2 to 4 wk. In the stress-period the number of live animals in the aquaria and in subsamples of 1 dm^2 from the sediment cores were counted every 2 to 5 days. In the recovery period we counted the live animals once every week. A 1-mm sieve was used to obtain the animals from the sediment subsamples.

The difference in number of live animals between the beginning and the end of an experiment was called the mortality (M) and was expressed as a percentage of the number of animals at the start of the experiment. In the laboratory experiments the numbers on the first and the last day were used to calculate the mortality, in the outdoor experiments the mean number of the first 4 days and last 2 wk were used. In addition, a mortality rate (R) was calculated. This is the decrease in numbers, expressed as a percentage of the number at the beginning of the experiment, per day during the stress-period. When the control showed a strong decline in numbers then the data were thought to be not reliable, and therefore rejected. When there was a continuous decrease during the recovery period, the mortality rate was calculated for a period up to and including 2 wk of the recovery period.

In the experiments with sediment cores the water content and salinity of the sediment in the cores were determined during the second week of the stress-period at intervals of 3 cm from the sediment surface. The water content (P) was measured as the loss of weight after drying at 65 °C for 3 days, and expressed as a percentage of the initial wet weight. By suspending (s g) sediment in (w g) tap water the salinity could be calculated from the chlorinity (C) measured with a chlorinity electrode connected to a Radiometer PHM 82, as follows:

$$S = (1.805 + 0.03) \times \left(\frac{w + P \cdot s}{P \cdot s}\right) \times C\,.$$

In the laboratory experiments at Yerseke the glycogen content of the animals was followed as an index of their condition (Ingle, 1949; Walne, 1970). The glycogen content was determined according to the method of Handel (1965) modified by De Zwaan & Zandee (1972a). The glycogen content was expressed as a percentage of the dry weight of the animals obtained after drying at 80 °C for 3 days.

RESULTS

CHANGES IN THE SALINITY AND WATER CONTENT OF THE SEDIMENT

Effects of prolonged dry periods (drainage) and the extra addition of fresh water (to simulate rain) were strongest in the uppermost 10 cm of the sediment (Fig. 3). The

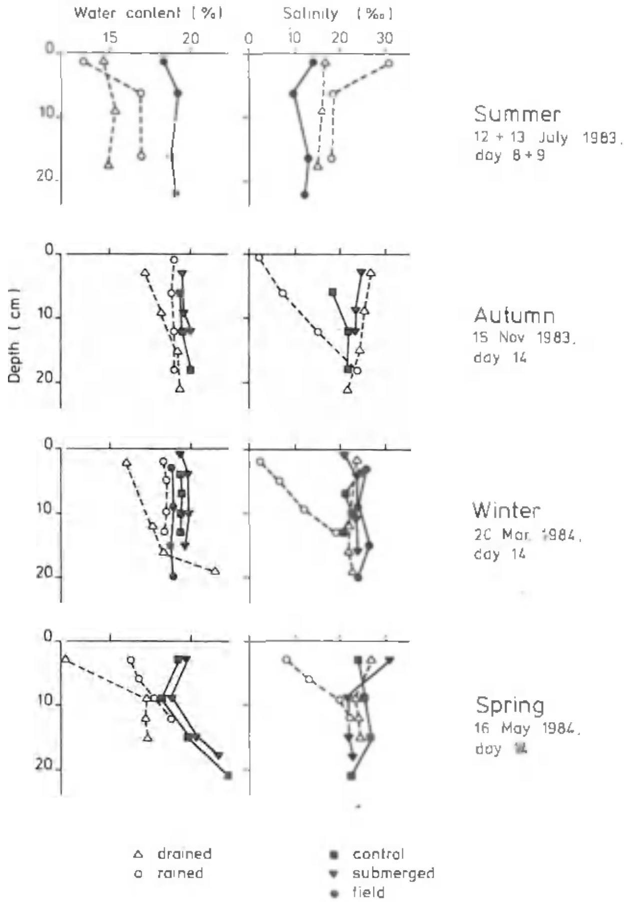

Fig. 3. Water content (% of the total wet weight) and salinity (‰ S) in the sediment after 8 to 14 days in the drained and submerged cores at Texel.

salinity and water content of the sediment in the submerged cores did not deviate from that in the control. For the summer a control was lacking and, therefore, a field sample was used as a control.

In summer the water content decreased and the salinity increased in both the drained and the rained cores (Fig. 3). These changes were probably caused by a strong evaporation, as a result of high temperatures (up to 30 °C) during the summer experiment (Table I). During the other seasons the changes in salinity and water content were different (Fig. 3). The decrease in the water content was greatest in the drained cores and, of course, less in the rained cores. In contrast to the summer, in the other seasons the salinity decreased in the rained cores, whereas in the drained cores it did not deviate from the control. This might be due to a minor evaporation and to the addition of fresh water in the rained cores.

THE LABORATORY EXPERIMENTS AT YERSEKE

Among the animals studied in the aquaria, strong differences in mortality in relation to the different species and seasons were found (Table II). Extended periods of submersion in stagnant water had hardly any effect; as a representative example the first (autumn) experiments are shown in Fig. 4. The mortality at the end of the experiments (*M*) and the mortality rate (*R*) were usually zero or negligible.

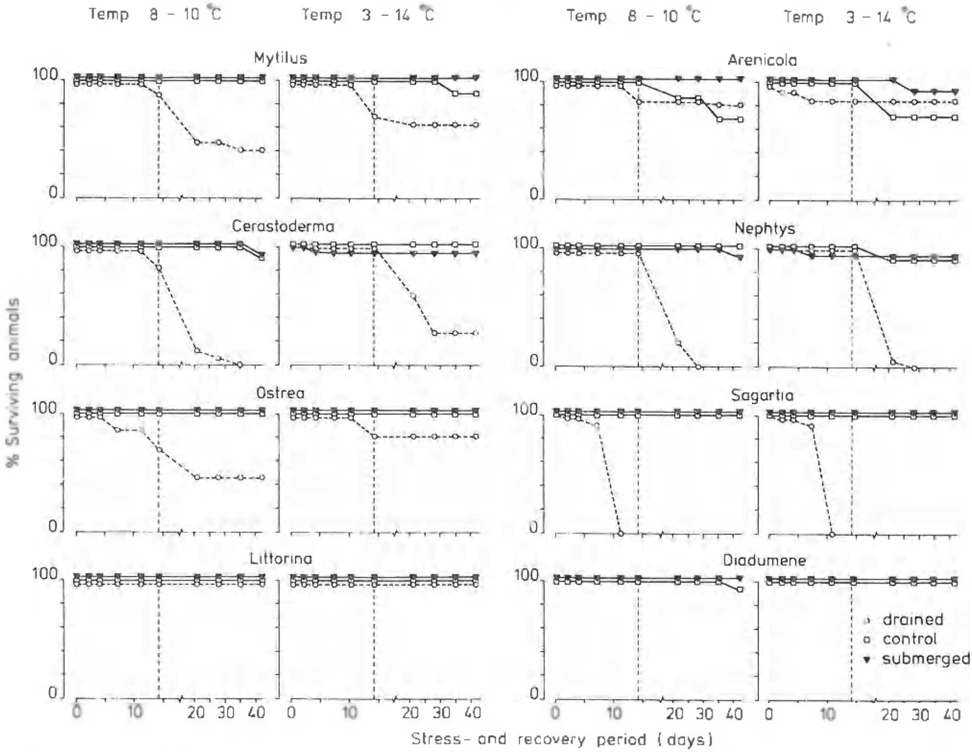

Fig. 4. Percentage of surviving animals in relation to the period of drainage or submersion in stagnant water during the autumn experiments in the laboratory at Yerseke: the vertical broken line indicates the end of the stress-period and the beginning of the recovery period.

Two weeks under dry conditions (drained), however, often caused a high mortality (Table II, Fig. 4). The decrease in number (percentage) started suddenly in most cases during the drainage period; the numbers dropped quickly to a minimum. The decrease in number often continued during the recovery period. The mortality and mortality rate of the animals in the room with constant temperature did not differ from that of the animals in the room with varying temperatures (Student's *t*-test, $P > 0.10$). Therefore the experiments in the two temperature rooms can be regarded as duplicates.

TABLE II

The mortality of the animals at the end of the experiments (M, %) and the mortality rate (R, %$\cdot$day^{-1}) as caused by drainage and submersion in stagnant water during the laboratory experiments

Season	Autumn								Winter							
Temperature (°C):	3 to 14				8 to 10				−3 to 9				2 to 4			
Stress-type	Drained		Submerged		Drained		Submerged		Drained		Submerged		Drained		Submerged	
M and R	M	R	M	R	M	R	M	R	M	R	M	R	M	R	M	R
Mytilus edulis L.	37	2.2	0	0	59	2.5	0	0	4	0.2	0	0				
Cerastoderma edule (L.)	73	2.0	6	0.2	100	4.2	0	0	13	0.4	0	0				
Ostrea edulis L.	20	1.4	0	0	54	2.6	0	0	11	0.5	32	0	9	0.4	0	0
Littorina littorea (L.)	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Arenicola marina (L.)	0	0	0	0	0	–	0	0								
Nephtys hombergi Savigny	100	4.5	0	0	100	3.8	7	0								
Sagartia troglodytes (Price)	100	9.1	0	0	100	9.1	0	0	100	6.4	12	0	46	2.0	0	0
Diadumene cincta Stephenson			0	0			0	0	100	4.8	0	0	100	4.8	15	1.7

Season	Spring								Summer							
Temperature (°C)	5 to 16				9 to 11				14 to 24				19 to 27			
Stress-type	Drained		Submerged		Drained		Submerged		Drained		Submerged		Drained		Submerged	
M and R:	M	R	M	R	M	R	M	R	M	R	M	R	M	R	M	R
Mytilus edulis	100	7.1			100	7.1			100	9.4			100	13.4		
Cerastoderma edule	100	7.1	0	0	100	7.1	29	–	100	25.0	18	2.6	100	25.0	64	3.0
Ostrea edulis	0	0			0	0			100	4.8			100	7.7		
Littorina littorea	0	0			0	0			20	1.6			27	1.9		
Arenicola marina	83	–			92	–			100	10.0			100	10.0		
Nephtys hombergi	100	7.1	0	0	100	7.1	0	0	100	14.3	0	0	100	14.3	0	0
Sagartia troglodytes	100	12.5			100	7.1			100	25.0			100	25.0		
Diadumene cincta	100	7.1	0	0	100	7.1	43	3.8	100	25.0	0	0	100	25.0	0	0

The anemones *Sagartia* and *Diadumene* and the polychaete *Nephtys* were the most susceptible to drainage (Table II). The most resistant to drainage was the periwinkle *Littorina*. The bivalves and the polychaete *Arenicola* were characterized by an intermediate mortality. The highest mortality rate was found in summer when most animals died within 4 to 10 days.

THE OUTDOOR EXPERIMENTS WITH SEDIMENT CORES AT TEXEL

The sediment cores, studied in an outdoor tank, were used to determine the reaction of a group of smaller organisms (Fig. 5; Table III). The species composition of the two stations sampled (high and low in the intertidal zone) differed strongly. Fig. 5 shows a representative example of the results during the first (summer) experiments. As in the laboratory experiments strong differences in the mortality of animals were found in relation to the different species and seasons.

A detrimental effect of extended periods of submersion in stagnant water could not be demonstrated (Table III). In contrast to this, most animals suffered heavily from extended dry periods. No difference was found between drained and rained samples

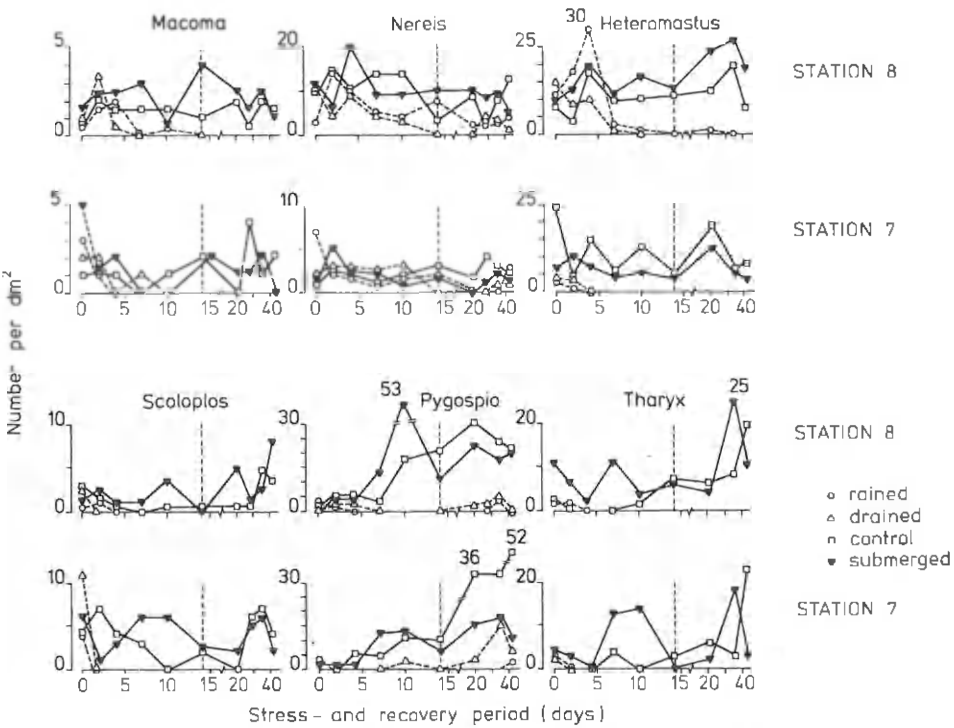

Fig. 5. The number of animals (per dm^2) in the drained, rained and submerged sediment cores during and after the stress-period in the outdoor experiments during summer at Texel: Station 7 lies high in the intertidal area, Station 8 is low; the vertical broken line indicates the end of the stress-period.

TABLE III

The mortality of the animals at the end of the experiments (M, %) and the mortality rate (R, % · day^{-1}) as caused by drainage (drained), precipitation during drainage (rained) and submersion in stagnant water (submerged) during the outdoor experiment with sediment cores.

Season and Station:	Summer, Station 8						Autumn, Station 8						Autumn, Station 7					
Temperature (°C):	10 to 30						−5 to 15						−5 to 15					
Stress-type	Drained		Rained		Submerged		Drained		Rained		Submerged		Drained		Rained		Submerged	
M and R	M	R	M	R	M	R	M	R	M	R	M	R	M	R	M	R	M	R
Macoma balthica (L.)	100	7.1	100	14.3	0	0	0	0	0	0	0	0						
Hydrobia ulvae (Pennant)													0	0	0	0	0	0
Nereis diversicolor O. F. Müller	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Heteromastus filiformis (Claparède)	100	10.0	100	4.8	0	0	100	4.8	97	4.8	0	0						
Scoloplos armiger (O. F. Müller)	100	50.0	100	25.0	0	0							100	5.0	100	4.2	0	0
Tharyx marioni (de St Joseph)	100	50.0	100	25.0	0	0	100	10.0	100	7.1	0	0						
Malacoceros fuliginosus (Claparède)													0	0	100	7.1	0	0

Season and Station	Winter, Station 7						Spring, Station 8						Spring, Station 7					
Temperature (°C):	−5 to 15						−1 to 18						4 to 15					
Stress-type	Drained		Rained		Submerged		Drained		Rained		Submerged		Drained		Rained		Submerged	
M and R	M	R	M	R	M	R	M	R	M	R	M	R	M	R	M	R	M	R
Macoma balthica							100	10.0			0	0						
Hydrobia ulvae	0	0	0	0	0	0												
Nereis diversicolor	0	0	0	0	0	0							0	0	64	1.9	0	0
Heteromastus filiformis							100	7.1	100	14.3	0	0						
Scoloplos armiger	100	4.8	100	6.7	0	0							100	7.1	100	11.1	0	0
Tharyx marioni											0	0						
Malacoceros fuliginosus	96	4.2	100	6.7	0	0							100	10.4	100	12.1	0	0

(Student's t-test, $P > 0.10$). Therefore, these two sets of data were regarded as duplicates.

The smaller polychaetes (*Heteromastus*, *Scoloplos*, *Tharyx*, *Malacoceros*) were particularly susceptible to drainage. The gastropod *Hydrobia* was the most resistant to drainage.

The mortality rate was highest during the summer experiments, some animals reaching 100% mortality within 2 days. During autumn and winter the mortality rates were so low that 100% mortality was not reached within 10 days of drainage.

SEASONAL INFLUENCES

For each season the mean mortality and mortality rate were calculated and their difference with other seasons was assessed (Fig. 6, Table IV). Clearly the mortality and mortality rate due to submersion in stagnant water were low in all seasons (Fig. 6). No significant differences in mortality were found between seasons (Table IV). Only the mortality and mortality rate in autumn were slightly lower than those in spring and summer.

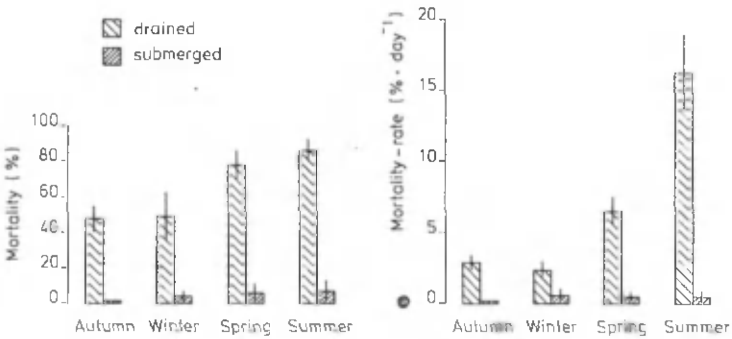

Fig. 6. The mortality and mortality rate of the animals during the experiments in the laboratory and in the outdoor tanks (means of all data in Tables II and III): data for drained animals include those for the rained cores: depicted are the mean and SE.

The mortality caused by extended periods of drainage showed a strong seasonal variation (Fig. 6). The mortality and mortality rate in both spring and summer were much higher than those in autumn and winter. The mortality rate during summer was, moreover, considerably higher than during spring.

THE GLYCOGEN CONTENT

The glycogen content of the animals varied throughout the year (Table V). The highest values, up to 30% in the bivalves, were found during summer and autumn, the lowest values during winter and spring. There was no significant difference in the glycogen content of the animals between summer and autumn, nor between winter and spring.

TABLE IV

Statistical significance of seasonal differences in total mortality (M) and mortality rate (R): *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$; ns, not significant, $P > 0.05$; Student's t-test.

Laboratory experiments at Yerseke

	Autumn	Winter	Spring	Summer	Drained samples
Autumn		ns	ns	**	Total mortality (M)
Winter	ns		*	***	
Spring	*	**		ns	
Summer	***	***	***		
	Mortality rate (R)				

	Autumn	Winter	Spring	Summer	Submerged samples
Autumn		*	*	*	Total mortality (M)
Winter	ns		ns	ns	
Spring	*	ns		ns	
Summer	**	ns	ns		
	Mortality rate (R)				

Outdoor experiments at Texel

	Autumn	Winter	Spring	Summer	Drained and rained samples
Autumn		ns	*	*	Total mortality (M)
Winter	ns		*	ns	
Spring	**	**		ns	
Summer	**	*	ns		
	Mortality rate (R)				

	Autumn	Winter	Spring	Summer	Submerged samples
Autumn		ns	*	*	Total mortality (M)
Winter	ns		*	ns	
Spring	**	**		ns	
Summer	**	*	ns		
	Mortality rate (R)				

An extended period of drainage or submersion did not alter the glycogen content of the animals compared with the control (as exemplified by the winter experiments in Fig. 7). The results were similar for all species and for all seasons.

DISCUSSION

Effects of submersion for 2 wk in stagnant water on benthic animals normally living on tidal flats were absent or negligible. In contrast to this we found a high mortality during extended periods of dry (ebb) conditions (drainage). Strong differences in the

TABLE V

Annual changes in the glycogen content of the animals at the beginning of the laboratory experiments (a) expressed as percentage of the highest glycogen content in the animals ($g \cdot g^{-1}$), and the statistical significance of the difference between seasons in the glycogen content (b); *, $P < 0.05$; ns, not significant, $P > 0.05$; Wilcoxon test; data of *Ostrea* omitted because only two results available.

(a) Glycogen content

	Autumn	Winter	Spring	Summer	Highest glycogen content
Mytilus edulis	64	33	5	100	0.249
Cerastoderma edule	8	13	8	100	0.241
Ostrea edule		79	100		0.278
Littorina littorea	100	64	49	45	0.074
Arenicola marina	100	65	85	50	0.026
Nephtys hombergi	100	38	50	48	0.052
Sagartia troglodytes	100	65	50	87	0.068
Diadumene cincta		41	32	100	0.034

(b) Difference between seasons in glycogen content

	Autumn	Winter	Spring	Summer
Autumn	–	*	*	ns
Winter		–	ns	*
Spring			–	*
Summer				–

degree and rate of mortality were found between species and seasons. Changes in the salinity and water content of the sediment were expected to contribute to the seasonal differences in mortality. The higher mortality during summer coincided with the higher salinities in the drained and rained cores. The upper limit for salinity tolerance for most marine benthic animals is >35 to 40‰ (Kinne, 1971; Wolff, 1973). In our summer experiments the salinity did not rise above 35‰ and is, therefore, not thought to be of great importance. Moreover, changes in the salinity and water content were similar during autumn, winter and spring. The seasonal changes in mortality of benthic animals are, therefore, unlikely to be influenced by changes in the salinity and water content of the sediment. Any influence of changes in salinity and water content on mortality is also questioned on the basis of the following facts. A decrease of the water content in the sediment was found in the drained cores, and to a lesser extent in the rained cores. A change of the salinity was found too, but only in the rained cores. No differences in the mortality between drained and rained animals were, however, found. The primary cause of this might be that the changes in water content and salinity were restricted to the upper 10 cm of the sediment, whereas, most of the animals would have retreated to the relatively undisturbed deeper layers of the sediment (Newell, 1976).

Other factors influencing the mortality of the animals might be the temperature and the condition (glycogen content) of the animals. Regarding the temperature, it proved

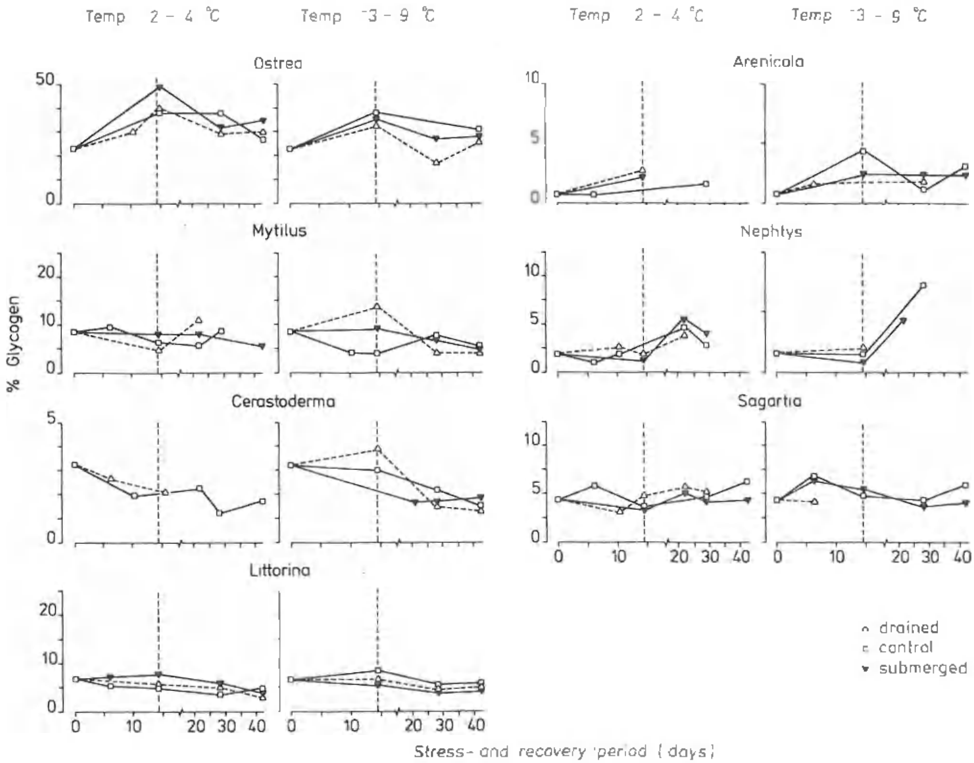

Fig. 7. The glycogen content (expressed as a percentage of the total dry weight) of the benthic animals in the laboratory experiments at Yerseke in relation to the period of drainage and submersion during the winter experiments: the vertical broken line indicates the end of the stress-period and the beginning of the recovery period.

not to be important whether it remained constant or varied 5 °C around the average as in our experiments. High temperatures are, however, likely to influence mortality. Boyden (1972) found a higher mortality in drained *Cerastoderma edule* with increasing temperature. The higher mortality and mortality rate during summer in our experiments (Fig. 6) may be explained by high temperatures: 30 °C during the outdoor experiments in summer and 27 °C in the laboratory experiments (Table I). A temperature of 25 to 30 °C is near the upper limit of thermal tolerance for marine benthic animals (Kristensen, 1958; Kennedy & Mihursky, 1972; Bayne, 1976; Ansell *et al.*, 1981). The low mortality in winter, might be explained by the low winter temperatures (3 to 5 °C); the metabolism of the animals will be very low at these temperatures, so that the animals can survive a longer stress-period (Newell, 1979). The temperature was moderate during spring and autumn. The mortality and mortality rate were, however, high in spring and low in autumn. Therefore, another factor must be involved in the high spring and low autumn mortalities; glycogen content could be this essential factor. Marine benthic animals can utilize their glycogen reserves during periods of stress (De Zwaan & Zandee, 1972a,b; Pandian, 1975; Dries & Theede, 1976; De Zwaan, 1977; Bayne *et al.*, 1982; Carr & Neff, 1984). Indeed, the glycogen content of the animals was low during spring and high during autumn (Table V).

Considering the above it is rather amazing that no decrease of the glycogen content was found during drainage (Fig. 7). A decrease of the glycogen has been found in several stressed benthic animals (Dales, 1958; De Zwaan & Zandee, 1972a; Dries & Theede, 1976; Newell & Bayne, 1980; Bayne *et al.*, 1982; Gäde, 1983; Akberali & Trueman, 1985). We, however, never found a decrease in any of the species studied during the four seasons.

Apart from differences between seasons, caused by differences in the condition of the animals and temperature, there are clearly species-dependent differences within a season. The smaller polychaetes and the (small) anemones, which are both without shells, suffered most from drainage (Tables II, III). The bivalves (with shell) and the bigger polychaetes had intermediate mortality rates. The gastropods, with a thick shell, have the highest resistance to drainage. As might have been expected morphological characteristics (size and shells) seem to dictate the species-dependent differences in mortality. Beside these morphological related differences our data suggest that the mortality rate of benthic animals during stress-periods is determined primarily by the temperature and the glycogen content. This means that benthic animals are more susceptible to disturbances when they have a low glycogen content and meet extreme (high or low) temperatures.

Acknowledgements

Thanks are due to Drs. A.B.J. Sepers, W.J. Wolff and A.W. Fortuin for critically reading the paper. This study was financed by the Ministry of Transport and Public Works, Rijkswaterstaat, Delta Department, Environmental Division.

References

Akberali, H.B. & E.R. Trueman, 1985. Effects of environmental stress on marine bivalve molluscs. *Adv. Mar. Biol.*, Vol. 22, pp. 101–198.

Ansell, A.D., P.R.O. Barnett, A. Bodoy & H. Massé, 1981. Upper temperature tolerance of some European molluscs. III. *Cardium glaucum* and *C. edule*. *Mar. Biol.*, Vol. 65, pp. 177–183.

Bayne, B.L., 1976. *Marine mussels: their ecology and physiology*. Cambridge University Press, Cambridge, 506 pp.

Bayne, B.L., A. Bubel, P.A. Gabbott, D.R. Livingstone, D.M. Lowe & M.N. Moore, 1982. Glycogen utilisation and gametogenesis in *Mytilus edulis* L. *Mar. Biol. Lett.*, Vol. 3, pp. 89–106.

Beukema, J.J., 1976. Biomass and species richness of the macrobenthic animals living on a tidal flat of the Dutch Wadden Sea. *Neth. J. Sea Res.*, Vol. 10, pp. 236–261.

Boyden, C.R., 1972. The behaviour, survival and respiration of the cockles *Cerastoderma edule* and *C. glaucum* in air. *J. Mar. Biol. Assoc. U.K.*, Vol. 52, pp. 661–680.

CARR, R.S. & J.M. NEFF, 1984. Field assessment of biochemical stress indices for the sandworm *Neanthes virens* (Sars). *Mar. Environ. Res.*, Vol. 14, pp. 267–280.

DALES, R.P., 1958. Survival of anaerobic periods by two intertidal polychaetes, *Arenicola marina* (L.) and *Owenia fusiformis* Delle Chiaje. *J. Mar. Biol. Assoc. U.K.*, Vol. 37, pp. 521–529.

DE ZWAAN, A., 1977. Anaerobic energy metabolism in bivalve molluscs. *Oceanogr. Mar. Biol. Annu. Rev.*, Vol. 15, pp. 103–187.

DE ZWAAN, A. & D.I. ZANDEE, 1972a. The utilization of glycogen and accumulation of some intermediates during anaerobiosis in *Mytilus edulis* L. *Comp. Biochem. Physiol.*, Vol. 43B, pp. 47–54.

DE ZWAAN, A. & D.I. ZANDEE, 1972b. Body distribution and seasonal changes in the glycogen content of the common sea mussel *Mytilus edulis*. *Comp. Biochem. Physiol.*, Vol. 43A, pp. 53–58.

DRIES, R.-R. & H. THEEDE, 1976. Stoffwechselintensität und Reservestoffabbau einiger mariner Muscheln bei herabgesetzter Sauerstoffsättigung des Mediums. *Kieler Meeresforsch.*, Sonderh. 3, pp. 37–48.

FORTUIN, A.W., 1981. Samenstelling, verspreiding, aantallen en biomassa van het macrozoobenthos in het Volkerak–Oosterschelde estuarium in de periode 1959 t/m 1976. *Delta Institute Reports, Yerseke*, No. 1981-6, 237 pp.

GÄDE, G., 1983. Energy metabolism of arthropods and mollusks during environmental and functional anaerobiosis. *J. Exp. Zool.*, Vol. 228, pp. 415–429.

HANDEL, E. VAN, 1965. Estimation of glycogen in small amounts of tissue. *Analyt. Biochem.*, Vol. 11, pp. 256–265.

INGLE, R.M., 1949. A comparative study of oyster condition. *Science*, Vol. 109, p. 593 only.

KENNEDY, V.S. & J.A. MIHURSKY, 1972. Upper temperature tolerances of some estuarine bivalves. *Chesapeake Sci.*, Vol. 12, pp. 193–204.

KINNE, O., 1971. Salinity, animals, invertebrates. In, *Marine ecology, Vol. I.2.*, edited by O. Kinne, Wiley Interscience, London, pp. 821–995.

KRISTENSEN, I., 1958. Differences in density and growth in a cockle population in the Dutch Wadden Sea. *Arch. Néerl. Zool.*, Vol. 12, pp. 351–453.

NEWELL, R.C., 1976. Adaptations to intertidal life. In, *Adaptation to environment*, edited by R.C. Newell, Butterworths, London, pp. 1–82.

NEWELL, R.C., 1979. *Biology of intertidal animals.* Marine Ecological Surveys Ltd., Faversham, 781 pp.

NEWELL, R.I.E. & B.L. BAYNE, 1980. Seasonal changes in the physiology, reproductive condition and carbohydrate content of the cockle *Cardium* (= *Cerastoderma*) *edule* (Bivalvia: Cardiidae). *Mar. Biol.*, Vol. 56, pp. 11–19.

PANDIAN, T.J., 1975. Mechanisms of heterotrophy. In, *Marine ecology, Vol. II.1.*, edited by O. Kinne, John Wiley & Sons, London, pp. 61–249.

WALNE, P.R., 1970. The seasonal variation of meat and glycogen content of seven populations of oysters *Ostrea edulis* L. and a review of the literature. *Fish. Invest., London*, Ser. II, Vol. 26, No. 3, 35 pp.

WOLFF, W.J., 1973. The estuary as a habitat. An analysis of data on the soft-bottom macrofauna of the estuarine area of the rivers Rhine, Meuse and Scheldt. *Zool. Verh.*, No. 126, 242 pp.