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EFFECT OF TIDAL SIMULATIONS AND ENTRAINMENT OF AN ENDOGENOUS TIDAL RHYTHM IN A NON-TIDAL POPULATION OF *GAMMARUS ZADDACHI*

by

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SUMMARY

The activity of *G. zaddachi* from a non-tidal environment (the brackish lake "De Putten", prov. North Holland, The Netherlands), has been studied in a current chamber in which tidal cycles can be simulated, in order to make a comparison with estuarine populations. The animals show a clear nocturnal activity pattern in stagnant water, which is preserved after transfer to running water, but with a reduced amplitude. Like in estuarine *G. zaddachi*, a very strong increase in swimming activity is provoked by a so-called complete high tide simulation, i.e. a decrease in current velocity, followed by a short period of standstill and a slow current in the opposite direction, and a rise in salinity (which resulted also in a rise in pressure of maximally 0.1 atm. due to pumping) and in temperature.

Contrary to what was concluded for estuarine populations, these non-tidal *G. zaddachi* show a much stronger response to changes in salinity than to changes in current. In fact, high tide simulations consisting of changes in salinity only result in an increase in activity, comparable with that at complete high tide simulations. A strong response is also noticed to a short increase in pressure of 0.3 and 0.5 atm., but the construction of the current chamber only allows tests of longer duration with an increase in pressure of 0.15 atm. This merely causes a small increase in swimming activity.

Remarkably enough, simulations in the light have the same effect on activity as simulations in the dark.

An (endogenous) circatidal activity rhythm with peak swimming at the expected time of simulated high tide is entrained in non-tidal *G. zaddachi* by subjecting them to a series of complete high tide simulations at tidal intervals. A series of high tide simulations, consisting of changes in current and salinity (but without changes in temperature) also entrains the rhythm, but neither changes in current nor changes in salinity are able to entrain the circatidal rhythm when applied alone (the same holds true for the small increases in pressure, due to pumping). The result of a combination of factors is therefore more than simply the sum of their individual effects.

The endogenous tidal rhythm does not show any sign of inhibition at expected high water in the light. Earlier fieldwork on the migration of estuarine *G. zaddachi* suggests that the tidal migrations of this species are confined to the dark. The activity of freshly caught estuarine *G. zaddachi* from the river Slack (France) shows a circatidal rhythm in the laboratory with equal peaks in the light and in the dark. The absence of tidal migrations during spring tides in daytime is therefore difficult to explain on the basis of the behaviour observed under laboratory conditions. Possibly the influence of light in the laboratory is different from that in the field, and further fieldwork will be necessary to determine the effect of light on emergence, height of swimming, etc., of estuarine *G. zaddachi.*

INTRODUCTION

Gammarus zaddachi Sexton, 1912 (Crustacea, Amphipoda) is a strongly euryhaline species, inhabiting the boreal brackish waters.

Field studies by Dennert et al. (1969) and Dennert & Van Maren (1974) in the estuary of the river Slack and by Girisch et al. (1974) in the river Dourduff — both small streams on the French Channel coast — have demonstrated that there is a complex relation between the up- and downstream migrations of *G. zaddachi* with the life cycle and with various environmental factors. All the year round there is a downstream (catadromous) migration ("organic drift"), mainly consisting of adults. It shows a nocturnal activity pattern with a peak some hours after sunset, as found in other gammarids (Müller, 1963). Only during the nocturnal spring tides, however, especially those of the autumn equinox, mass migration takes place in upriver (anadromous) direction by utilizing the reversal of the current. In this upstream migration both adults and juveniles participate. (Up- and downstream migration is thus meant here in a topographic sense.)

Although *G. zaddachi* utilize the current for migration, they are not carried away passively by the current. Dennert et al. (1969) demonstrated that the migration is an active process, depending on the physiological state of the animals. *G. zaddachi* physiologically in catadromous phase continue to move downstream, irrespective of the actual direction of the current. Likewise, animals in anadromous phase continue moving in upstream direction.

To establish which environmental factor or combination of factors is triggering the upstream migration of *G. zaddachi* and *G. chevreuxi,* Girisch & Dennert (1975) started simulation experiments with a current chamber. They could simulate natural high tides completely or partially by regulating important tidal factors like temperature, salinity, current speed and current direction. To compare the results with those of the previous fieldwork, they sampled their experimental populations from the same streams, the rivers Slack and Dourduff.

Highest activity was found when a complete high tide was simulated (i.e. a decrease of the current, followed by a standstill and a slow current in the opposite direction, combined with a rise in temperature and salinity). By leaving out the change in one of the factors, a less pronounced increase in activity was obtained. The main factor causing an increase in activity seemed to be the change in current velocity and direction. The sole influence of a change in salinity was also considerable. Contrary to a long lasting rise in temperature of 6 °C during several days, a quick rise in temperature (of tidal amplitude and duration) did not influence the behaviour of *G. zaddachi.*

In order to control whether this typical behaviour was restricted to estuarine populations, Girisch & Dennert (1975) did also some experiments with a population from the North Sea Canal (The Netherlands), a stagnant brackish water. They did not find large differences between the estuarine and the non-tidal populations concerning the effect of the above-mentioned factors.

Another factor, which might influence the activity of *G. zaddachi* and which was not examined by Girisch & Dennert in their simulation experiments is the change in hydrostatic pressure occurring at high tide. Several marine and intertidal animals show responses to changes in pressure. This has been demonstrated for the pycnogonid *Nymphon gracile* (cf. Morgan et al., 1964), the amphipod *Corophium volutator* (cf. Morgan, 1965), the swimming crab *Macropipus (=Portunus) holsatus* (cf. Morgan, 1967) and the amphipod *Synchelidium* sp. (cf. Enright, 1961, 1962), all species using the tides for migration. In nature, estuarine populations of *G. zaddachi* are subjected to pressure cycles of considerable amplitude, so the influence of this factor on the behaviour of this species is certainly worth testing.

It was shown by Girisch & Dennert (1975) that *G. zaddachi* presents a well-developed nocturnal activity pattern in the laboratory, corresponding with the nocturnal migratory activity observed in the field studies. Besides diel periodicity, many marine organisms show a periodicity in activity corresponding with the lunar day (24.8 h) or the tidal interval (12.4 h). Often such an activity pattern persists when the organisms are kept isolated from tidal influence in the laboratory and thus is apparently endogenous. The existence of such endogenous rhythms in isolation from the main environmental cycles is usually explained by a timing mechanism within the organism, the so-called biological clock. Because the period of such endogenous rhythms often slightly differs under constant conditions from the exact geophysical period, they are usually called circatidal or circalunadian rhythms (Palmer, 1973). Dieleman (1977) found such a circatidal rhythm in freshly sampled estuarine *G. zaddachi.* This activity rhythm could partially explain the mechanism of anadromous migration. Especially during spring tides, estuarine *G. zaddachi* in the laboratory, isolated from tidal influence, show a peak in swimming activity at the expected time of high water. They are then loosing their normally positive rheotaxis [1]) and become more or less indifferent to the current. Under natural conditions such a behaviour, combined with the triggering effect of changes in current and salinity described above, will result in anadromous migration during the reversal of the current at spring tides.

Such endogenous rhythms are entrained (synchronized) by some tidal factor. Pressure cycles of tidal periodicity can entrain a persistent tidal rhythm in the isopod *Eurydice pulchra* (cf. Jones & Naylor, 1970). In the crab *Carcinus maenas,* cycles in pressure (Naylor & Atkinson, 1972), in temperature (Williams & Naylor, 1969) and in

[1]) According to Arnold (1974), taxes are directed reactions (opposite to kineses). With a single source of stimulation the body is orientated in line with the source and movements towards or away from it are described as positive or negative.

salinity (Taylor & Naylor, 1977) are effective synchronizers. Wave action has been shown to be effective as well, viz. in the isopod *Excirolana chiltoni* (cf. Klapow, 1972).

The aim of the present study can be summarized as follows. In the first place, it will be tested more extensively whether non-tidal *G. zaddachi*, living for many years in stagnant brackish water, do react in the same way on tidal simulations as estuarine *G. zaddachi*. This was concluded by Girisch & Dennert (1975) on the basis of few experiments only. Like they did for estuarine populations, we will try to establish which tidal factor (or combination of factors) is giving the most pronounced triggering effect on the swimming activity of non-tidal *G. zaddachi*. Furthermore it will be tested whether treatment by a series of tidal simulations at tidal intervals is able to induce a persistent tidal activity rhythm in non-tidal *G. zaddachi*, and if so, which tidal component (or combination of components) is the effective synchronizer.

MATERIAL AND METHODS

The experimental populations of *G. zaddachi* were sampled with a dip net in a small brackish lake, "De Putten" near Camperduin (prov. of North Holland, The Netherlands), which has been secluded from the sea for over a century. The present lake was created during a reconstruction of the Hondsbossche Zeewering, which is a big dike closing a gap in the dunes in the neighbourhood of the city of Alkmaar, in the years 1865-1870 (pers. comm. of Mr. Zuidweg of the Water Board Hoogheemraadschap Noordhollands Noorderkwartier). Thus these gammarids have been living without tidal influences for many generations (their life cycle lasts about one year). The animals were transported in lake water (specific conductivity about 10^4 μS) to the Institute of Taxonomic Zoology in Amsterdam. The experimental populations consisted mainly of adults, with some juveniles which were always larger than 5 mm. The sex ratio was approximately 1.

The gammarids were put in the same current chamber as used by Girisch & Dennert (1975); see their paper for a detailed description. A second current chamber of the same type was now available and so parallel experiments were possible.

An improvement concerning the regulation of the light/dark cycle was made. No normal daylight could penetrate into the current chamber. Two 100 Watt daylight spectrum lamps, illuminating the current chamber during the day (with a maximum light intensity of about 900 Lux at the water surface), were connected with a light sensitive cell outdoors. This light cell, connected with a dimmer, was regulating the light/dark periods, following the normal light/dark cycles in nature, with a simulated dusk and dawn.

To increase salinity in the current chamber during the simulation experiments, seawater was used from the North Sea at IJmuiden or Camperduin (The Netherlands), and to decrease salinity normal tap-water was used. Due to the pumping of sea- or tap-water, pressure in the chambers was increased with maximally 0.1 atm.

In an observation period of 5 minutes at intervals of 1 hour or less, the activity was measured by counting the animals crossing a vertical reference line on the inner chamber wall. During the activity peaks at high tide simulations, often counts of 2.5 minutes were made at short intervals; the number was then doubled in order to estimate the activity in 5 minutes. In the first experiments counts were made by direct visual observation, using a narrow beam of dim red light in the dark period. Later a video camera suitable for daylight as well as for weak infrared light was used. Time lapse recording on video tape allowed counting of activity at any desired time (a digital electric clock, covered by an infrared filter and placed in the middle part of the current chamber, was always visible on the TV screen). The use of these different kinds of light in the dark period in the two methods of observation did not exert any influence on the activity pattern of *G. zaddachi*.

The current chamber was labelled according to the normal set of the current (see below) in an up- and downstream direction. Animals moving in these labelled directions are referred to as up- and downstreamers, respectively, irrespective of the actual current direction, and were counted separately. This implies, that during the reversal of the current at simulated high water, animals actively swimming against the current are still called downstreamers, because they move in the direction labelled downstream.

The current velocity is expressed in the voltage (V) of the motor driving the propellor which is generating the water current. A table for the relation between voltage and current velocity is given by Girisch & Dennert (1975, table II). Their table clearly shows that the same voltage results in a far slower current when the current direction is switched from the normal downstream in the upstream direction (for instance, 4 V in downstream direction corresponds with about 6-7 cm/sec, whereas 4 V in upstream direction corresponds with only 2.5-3 cm/sec).

Salinity is expressed in specific conductivity (μS). During the experiments the pH of the water was always close to 8, with some minor fluctuations. The water was always practically saturated with oxygen. Food, the common weed *Stellaria media* Villars and/or decaying leaves, was always available. Gravel and pebbles (2-35 mm) from the river Slack were used as a substrate in a layer of about 3 cm, thus shelter was sufficiently available.

RESULTS

In order to check if the experiments were influenced by too high population densities, both a population of 100 and one of 200 *G. zaddachi* were put in the two current chambers under the same conditions, corresponding with lake temperature and salinity. In the case of an overpopulation a rise in activity during daytime is to be expected, as part of the population will not be able to find shelter and will keep swimming around in search for it.

Fig. 1 shows the normal diel activity pattern of these two populations, both without current (V = 0, fig. 1a) and after the current has been started (V = 4, fig. 1b). The population of 200 gammarids does not show a relatively higher activity at daytime compared with the population of 100, thus we can assume that a population density of 200 *G. zaddachi* is still acceptable.

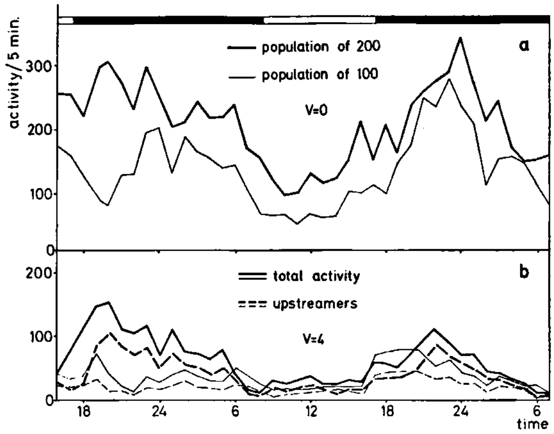

Fig. 1. (a) Normal diel activity pattern of *G. zaddachi* at two population densities in stagnant water (V = 0). Bars indicate light and dark. (Temperature 6 °C, specific conductivity 10^4 μS.)

(b) Diel activity pattern of the same populations in running water (V = 4). The current was started 7 days before. The animals were caught on 4 and 6 January 1977 and were put in the current chamber on 12 January. Survival: 83 and 45 animals were still alive on 4 February (of 200 and 100, respectively). Note that the upstreamers constitute the major part of the total activity. (Temperature 6 °C, specific conductivity 10^4 μS.)

The transition from stagnant to running water gave a disturbance, but after some days the diel activity pattern was back to normal (though the total activity was considerably lower in running water, cf. figs. 1a and 1b). Therefore, before starting an experiment in which the tidal environment was simulated, the animals were acclimated for some days to the new circumstances, corresponding with lake temperature (except for the winter months, when a temperature of some °C above lake temperature was used) and with the current (V = 4) in the downstream labelled direction. The specific conductivity of the water was about 7000 μS, obtained by diluting the lake water slightly with tap-water. These circumstances are comparable with ebb-tide conditions, though in the estuary at low tide the conductivity can be much lower (about 1000 μS). No further dilution was made, however, to avoid high mortality rates. In the water used here, the major part of the activity consists of upstream movements (fig. 1b). In fresh water (specific conductivity 600 μS) downstream movements dominate when *G. zaddachi* originating from stagnant brackish water are put in the current chamber (Girisch & Dennert, 1975).

The effect of complete high tide simulations

A complete high tide simulation means a step by step decrease of the current velocity till a standstill is reached, reversal of the current with a low current velocity in the upstream labelled direction, while at the same time a rise in temperature ($\Delta T = 4$ °C) and salinity (to about 30,000 µS) is accomplished, followed after some time by a gradual decrease of the current and after a short period of standstill, a reversal of the current back to the original direction, combined with a decrease in temperature and salinity, so that the ebb-tide conditions are reached again. The whole procedure takes about 1.5 to 2 hours.

Fig. 2 shows the activity pattern of *G. zaddachi* during the last 2 complete high tide simulations of a series of 21 at tidal intervals. The activity pattern during the following two days under constant conditions is shown in fig. 3. In our experiments, constant conditions always mean: ebb-tide conditions with a normal light/dark regime. During the high tide simulation, activity increases clearly, both when the current is reduced and when salinity is raised, and drops sharply back to a quite low level when the ebb-tide situation is reached again (fig. 2).

In the following observation period under constant conditions (fig. 3) an obvious tidal rhythm is visible: activity peaks appear at the expected time of high water (indicated by arrows in the figure), both in the light and in the dark. Only the first peak is somewhat retarded and shifted to the dark period. Clearly, this treatment is able to entrain an endogenous tidal rhythm in these non-tidal *G. zaddachi*, with a period very close to 12.4 hours.

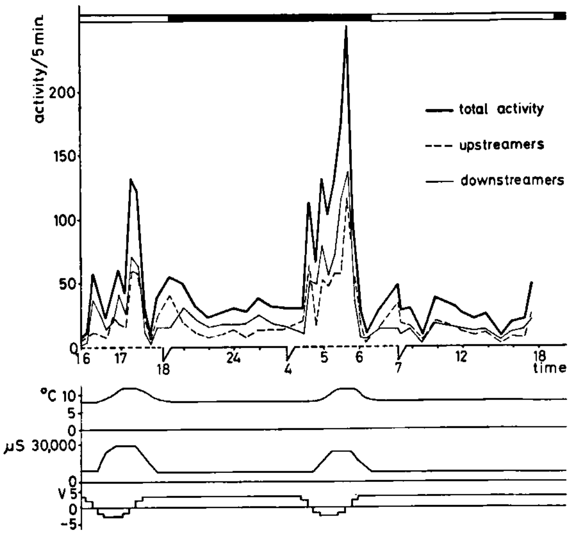

Fig. 2. Activity pattern of *G. zaddachi* during the last 2, of a series of 21, complete high tide simulations. Note the different time scale during these simulations in order to represent the counts at short-term intervals. Bars indicate light and dark. Continuation given in fig. 3.

The normal nocturnal activity pattern before treatment (see fig. 1) has completely disappeared and has been replaced by a pattern with equal activity peaks in the light and in the dark, both during the series of simulations as well as in the subsequent period under constant conditions. (N.B. the inequality of the peaks during the simulations in the light and in the dark in fig. 2 is exceptional.)

The presence of a rhythmic component with a

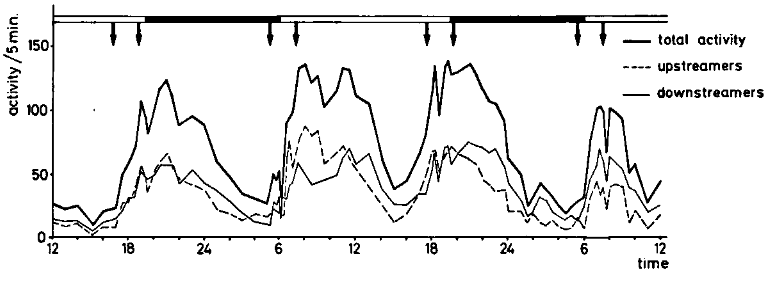

Fig. 3. Activity pattern during the two days under constant conditions (except for light and dark, indicated by bars) following the last complete high tide simulation in fig. 2. Arrows indicate the expected time limits of simulated high tide. (Temperature 8 °C, specific conductivity 7500 µS, V = 4.) The animals were caught on 8 March 1977 and 150 specimens were put in the current chamber on 9 March. Survival: 103 specimens were still alive at the end of the observations on 4 April.

period of approximately 12.4 hours is confirmed by subjecting the data to spectral analysis, a technique which can be used to determine the most prominent cycle(s) in empirical time series. The subprogram SPECTRAL of the Statistical Package for the Social Sciences (Aarons & Reagan, 1977) was used here. For each hour of the period under constant conditions mean activity per time unit (minute) is computed and these mean activity values are used in the analysis. The program computes autocovariance and -correlation as well as a power spectrum, indicating the strength of the various periodic tendencies in the data (on the basis of Fourier methods). As our series of observations are often too short to give a detailed picture of this power spectrum in the range of periodicities we are interested in (roughly from 12 to 24 hours), only the autocorrelation diagrams are used here. The autocorrelation coefficient presents the relation between activity at a certain hour (say *n*) and the activity after a certain fixed time interval (say time lag *L*), with $n = 1, 2, \ldots, N - L$, in which *N* is the total number of hourly activity values under constant conditions. The coefficient is computed for the whole range of time lags we are interested in (generally $L = 0, 1, 2, \ldots, 25$ h was used). The resulting picture of autocorrelation coefficients at different time lags gives a good indication about the strongest periodic tendencies in the observations. When activity shows a recurrent pattern with a period of *X* hours, time lags of $L = X$ will result in positive correlation coefficients. The coefficients plotted in fig. 4 (concerning the activity data presented in fig. 3) clearly show a basic periodicity close to the tidal one.

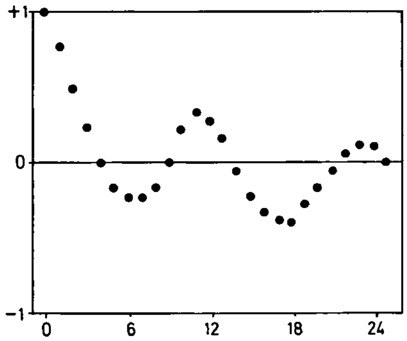

Fig. 4. Analysis of periodicity in the data presented in fig. 3, concerning activity under constant conditions after treatment by 21 high tide simulations at tidal intervals. Autocorrelation coefficients between mean hourly activity values at different time lags are plotted. For further explanation see the main text; 104 observations in 63 hours were used.

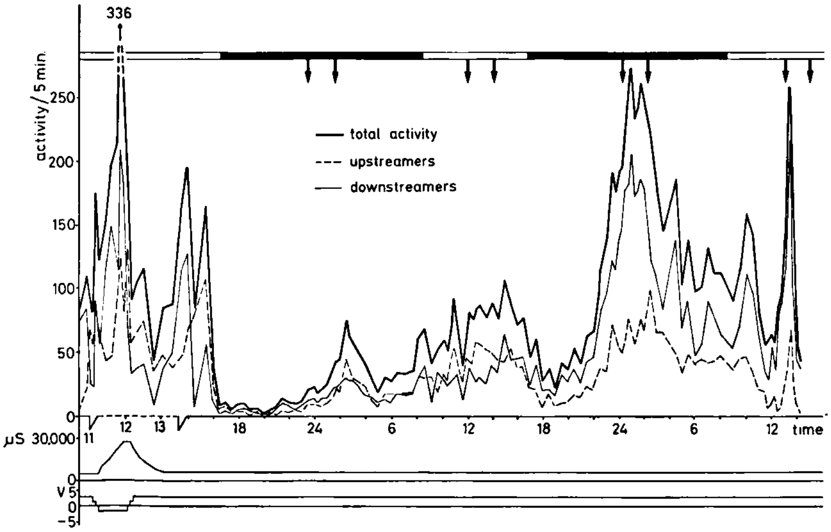

Fig. 5. Activity pattern of *G. zaddachi* during the last of a series of 15 high tide simulations (consisting of changes in current velocity and direction and in salinity, but unlike the treatment in fig. 2, without changes in temperature) and during the next two days under constant conditions. Bars indicate light and dark; arrows indicate the expected time limits of simulated high tide. On 18 November 1976, 234 animals were put in the current chamber (temperature 11 °C); 166 were still alive at the end of the observation period on 11 December 1976.

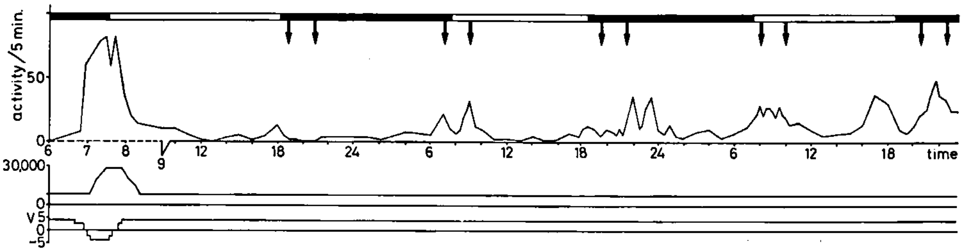

Fig. 6. As in fig. 5, but here the total activity during the last of a series of 16 high tide simulations (with changes in current and in salinity) is shown together with the following period of 2.5 days under constant conditions. Bars indicate light and dark; arrows indicate the expected time limits of simulated high tide. The animals were caught on 7 February 1977 and 125 specimens were put in the current chamber on 11 February (temperature only 6 °C). Survival: 61 animals still alive on 7 March 1977.

The effect of complete high tide simulations without a rise in temperature

The conditions during these experiments were almost the same as in the previous one, except that there was no rise in temperature during the high tide simulations. Figs. 5 and 6 show the activity patterns during the last of a series of 15 (fig. 5) or 16 (fig. 6) of such partial high tide simulations at tidal intervals and the following two days under constant conditions. The gammarids react with relatively the same intensity at these high tide simulations compared with those including a rise in temperature. Although only a small activity peak is observed at the first expected time of high water under constant conditions in both series, the next peaks clearly appear at the expected times of the high tide simulations.

In spite of the clear activity peaks at the expected times of high water under constant conditions in fig. 5, the correlation between activity values at different time lags (fig. 7a) does not indicate a circatidal activity rhythm as clearly as in the previous experiment (fig. 4). This is apparently due to the unequal height of the activity peaks of the two days under constant conditions. When the activity is divided by the daily mean (counted from sunset to sunset), a circatidal activity rhythm is clearly visible (fig. 7b).

The reason why fig. 6 does not give such a pronounced picture as fig. 5 is probably due to the relatively low activity, possibly caused by the high mortality rate (61 animals of the initial 125 were still alive after the experiment) and by the rather low temperature (T = 5-6 °C) compared with the experiment in fig. 5 (T = 10-11 °C). Despite of

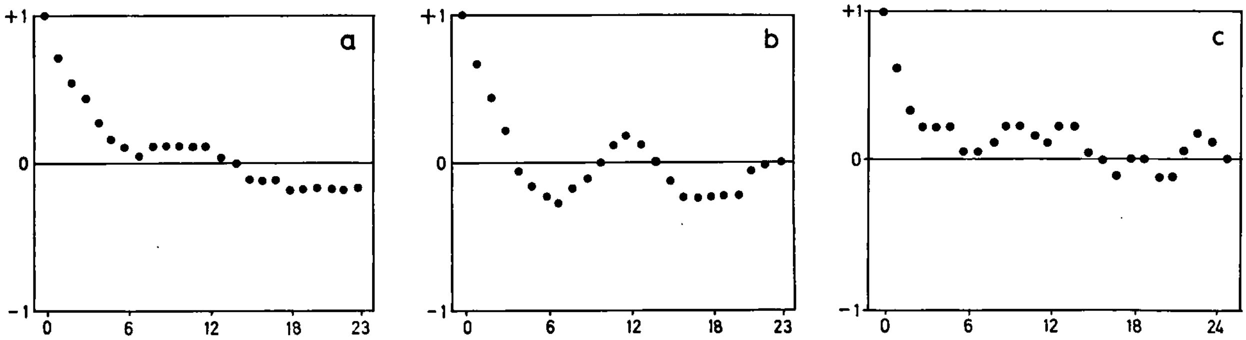

Fig. 7. (a) Analysis of periodicity in the activity data of the first two days under constant conditions (after treatment by a series of tidal cycles consisting of changes in salinity and current velocity and direction), of the experiment in fig. 5; 97 observations in 49 hours were used. For further explanation see the main text.

(b) Do., concerning the activity data used in fig. 7a after transformation in order to mask the unequal peak height on the successive days, by dividing the mean value for each hour by the mean diel activity (counted from sunset to sunset). The circatidal periodicity is now much more clearly visible than in fig. 7a.

(c) As (a), concerning the activity data under constant conditions of the experiment from fig. 6; 101 observations in 64 hours were used.

this, the autocorrelation diagram still shows a scattered relative maximum around 12.4 hours (fig. 7c), indicating the presence of a circatidal rhythm.

The effect of changes in current velocity and direction only

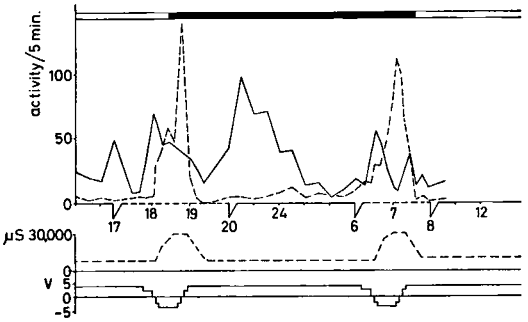

Fig. 8. Different effects on activity pattern of simulations consisting of changes in current only (————) and of changes in both current and salinity (— — — —). In the latter case activity is strictly limited to the hours of simulated high tide, unlike the former case, where also a marked activity peak following sunset is observed. The data originate from the parallel experiments of figs. 6 and 10. Because the experiment with current changes only started with delay due to technical problems, the graph shows the 7th and 8th cycle in current, compared with the 13th and 14th cycle in the parallel experiment. For further details on experimental conditions and populations see figs. 6 and 10. Bars indicate light and dark.

Two experiments ran parallel with those presented in figs. 2 and 3 and in fig. 6, but here variations in both temperature and salinity were left out. In this way the sole influence of changes in current velocity and direction during simulated high tide could be observed. In fig. 8 two of these simulations are shown (uninterrupted line) together with the result of the simultaneous complete high tide simulations from the experiment of fig. 6 (interrupted line). Although there is always a response to the change in current, the response is quite variable and far less than that to the complete high tide simulations in the parallel experiment. Moreover, in these experiments with changes in current only, the normal nocturnal activity pattern is still dominating, whereas in the parallel experiment (with changes in both current and salinity) the activity runs completely in phase with the imposed artificial tidal cycle.

Figs. 9 and 10 (uninterrupted lines) show the activity pattern during the last of a series of 21 and of 10 of such tidal cycles with changes in current only, as well as the activity during the following period under constant conditions. Obviously there is a normal nocturnal activity pattern during the two days following the last simulation.

In both cases activity is quite low, possibly due

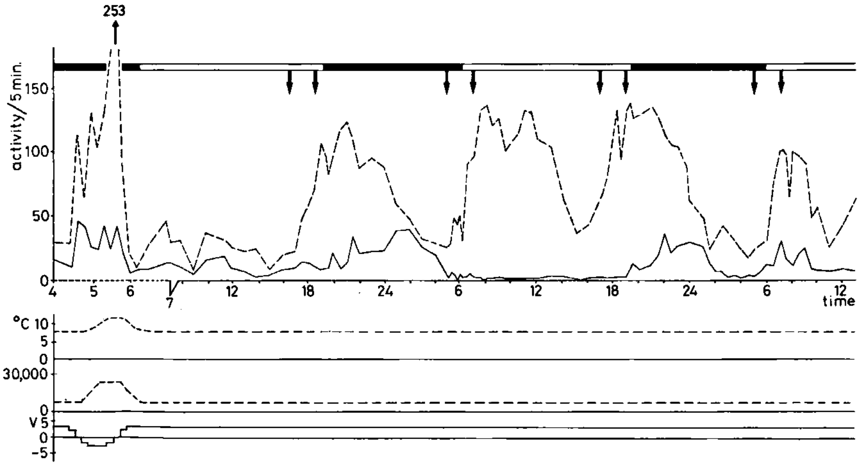

Fig. 9. Activity pattern of *G. zaddachi* during the last of a series of 21 high tide simulations consisting of changes in current velocity and direction only, and during the following period under constant conditions (uninterrupted line). For comparison the results of the parallel experiment with a series of 21 complete high tide simulations, as shown in figs. 2 and 3, are given as well (interrupted line). Bars indicate light and dark; arrows indicate the expected time limits of simulated high tide. The animals were caught on 8 March 1977 and 150 specimens were put in the current chamber on 9 March (temperature 8 °C). Survival: 55 specimens still alive on 4 April 1977.

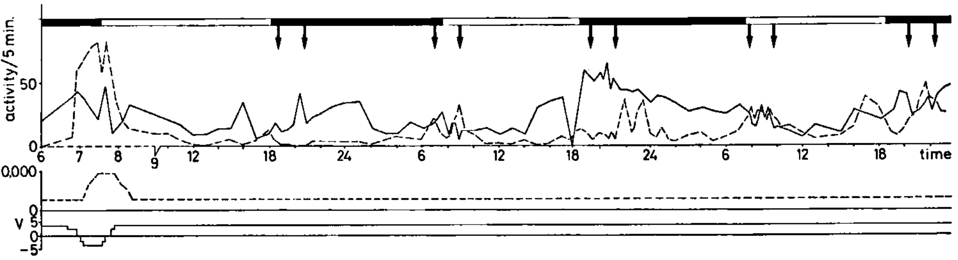

Fig. 10. Like fig. 9, but here the activity pattern during the last of a series of 10 high tide simulations with changes in current velocity and direction only, and during the following period of 2.5 days under constant conditions is shown (uninterrupted line). The interrupted line shows the results of the parallel experiment with a series of 16 high tide simulations (with changes in current and salinity, but without a rise in temperature) as plotted in fig. 6. Bars indicate light and dark; arrows indicate the expected time limits of simulated high tide. The animals were caught on 7 February 1977 and 125 specimens were put in the current chamber on 11 February (temperatue 6 °C). Survival: 38 specimens were still alive on 7 March 1977.

to the high mortality rate: 55 animals (of a population of 150) were still alive after the experiment of fig. 9 and 38 (of a population of 125) at the end of the experiment of fig. 10. The high mortality rate in these experiments compared with those in the experiments of figs. 3, 5 and 6 corresponds with the results of Dennert & Van Maren (1974) and Girisch et al. (1974), namely that a higher or periodically raised salinity has a positive influence on survival. Possibly also the fact that here no daily refreshing of the water occurs, unlike in the experiments with simulations including a change in salinity, influences mortality.

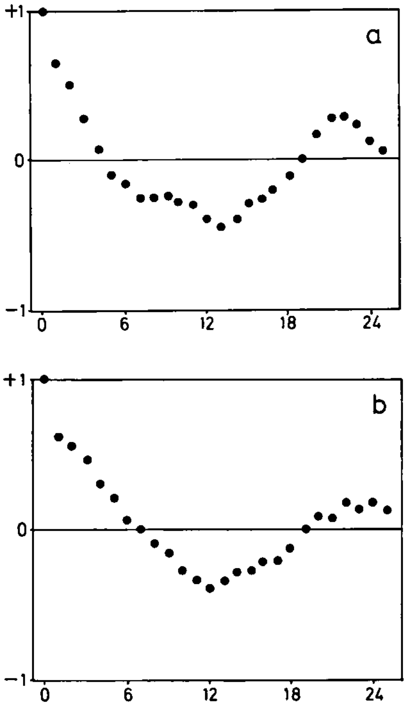

Fig. 11. (a) Analysis of periodicity in the activity data of the period under constant conditions, after treatment by tidal cycles consisting of changes in current only, of the experiment from fig. 9; 96 observations in 63 hours were used. (b) Do., concerning the activity data under constant conditions of the experiment from fig. 10; 94 observations in 63 hours were used.

For the period under constant conditions in figs. 9 and 10 the autocorrelation diagrams (figs. 11a and 11b, respectively) only show a maximum around 24 hours, not around 12.4 hours. Thus after treatment by tidal cycles, consisting only of these changes in current, a normal diel rhythmic pattern in activity is found, and not a tidal one.

The effect of changes in salinity only

From figs. 12, 13, 15 and 16 it is clear, that a rise in salinity has a strong influence on swimming activity. In fact, such a partial high tide simulation seems to provoke an increase in activity of the same magnitude as does a complete high tide simulation. Remarkable is, that often a second peak in activity occurs when salinity is decreasing again, or just after the ebb-tide values have been attained. This corresponds with the results of Taylor & Naylor (1977) with the crab *Carcinus maenas,* that every change in salinity induces an activity peak.

Striking was the rheotactic behaviour during these second activity peaks in the experiment of fig. 13, shown in detail in fig. 16. For the whole series of 15 simulations a domination of upstream movements was observed when high water oc-

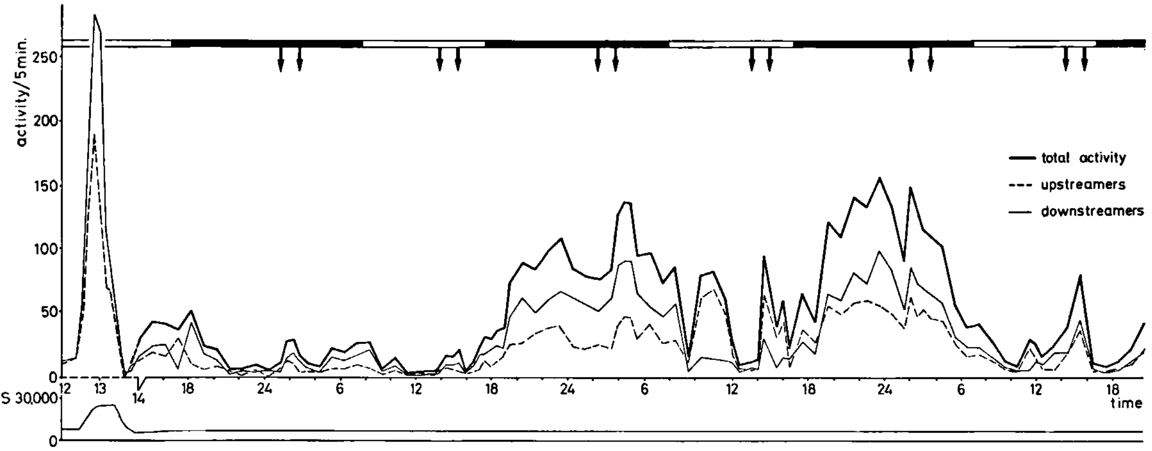

Fig. 12. Activity pattern of *G. zaddachi* during the last of a series of 14 high tide simulations, consisting of changes in salinity only, and during the following period of 3 days under constant conditions. Bars indicate light and dark; arrows indicate the expected time limits of simulated high tide. The animals were caught on 19 October 1978 and 200 specimens were put in the current chamber on 27 October (temperature 10 °C, V = 4). Survival: 125 animals were still alive on 14 November 1978. In view of the high activity during the simulation no total activity is given in that period.

curred in the light, whereas in the dark downstream swimming was dominating during these second activity peaks. This suggests that light could play an important role in the orientation towards the current. However, this phenomenon was not observed as clearly in the other series (figs. 12 and 15), where downstream swimming always dominated during the second activity peaks. During the first peak, at the increase of salinity, the number of up- and downstreamers is more or less equal, or the upstreamers are more numerous, which means in these experiments the animals actively swimming against the current. An exception is found in fig. 12, where downstreamers dominate during the activity peak coinciding with the rise in salinity. Active swimming against the current generally also dominates in the simulations including a reversal of the current, viz. downstreamers are more numerous during the period of current reversal, see for instance figs. 2 and 5. This was also found by Girisch & Dennert (1975).

The activity during the last cycle in salinity (of series of 14 and 15, respectively) and during the following period under constant conditions is shown in figs. 12 and 13. Obviously this treatment does not result in the entrainment of a persistent tidal rhythm and nocturnal activity dominates, though sometimes vague activity peaks occur at the expected time of high water, but these peaks are not at all convincing. Analysis of periodicity in the

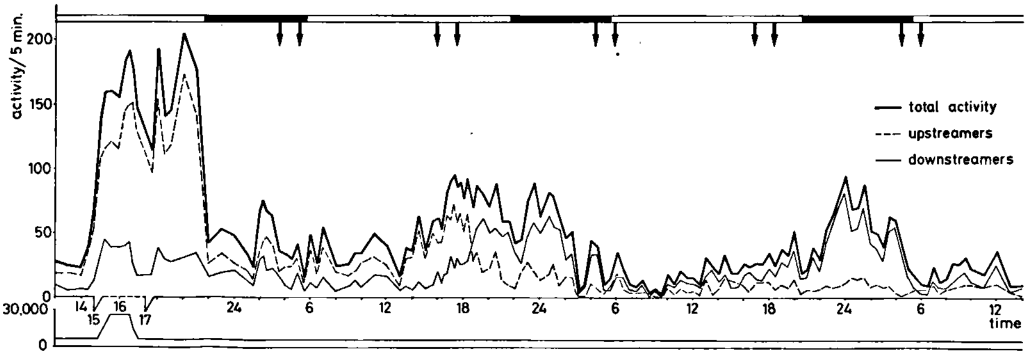

Fig. 13. Like fig. 12, but here the activity pattern during the last of a series of 15 high tide simulations (with changes in salinity only) and during the following period under constant conditions is shown. Bars indicate light and dark; arrows indicate the expected time limits of simulated high tide. The animals were caught on 24 April 1979 and 100 specimens were put in the current chamber on 28 April (temperature 12 °C, V = 4). Survival: 60 specimens were still alive on 15 May 1979.

activity data of the period under constant conditions does not give indications of the presence of a circatidal rhythm either (figs. 14a and 14b, respectively).

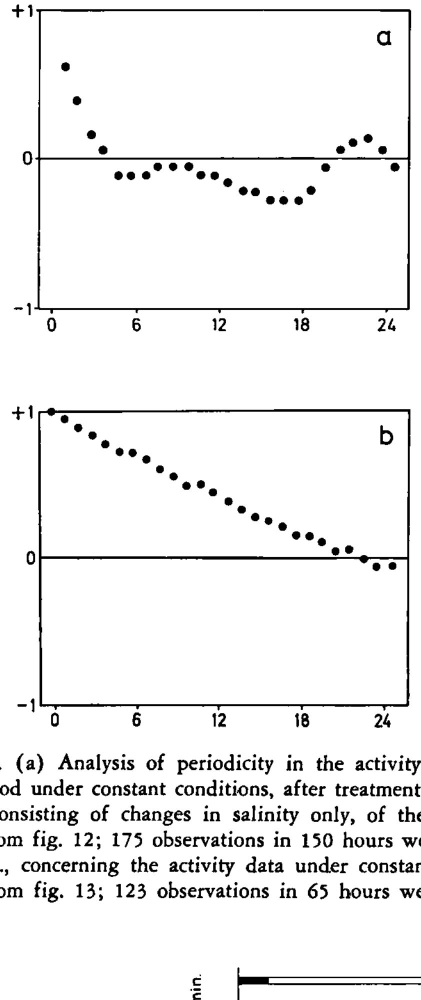

Fig. 14. (a) Analysis of periodicity in the activity data of the period under constant conditions, after treatment by tidal cycles consisting of changes in salinity only, of the experiment from fig. 12; 175 observations in 150 hours were used. (b) Do., concerning the activity data under constant conditions from fig. 13; 123 observations in 65 hours were used.

The effect of hydrostatic pressure only

In all experiments including changes in salinity, pumping always caused a rise in pressure of maximally 0.1 atm. Therefore an experiment was started in order to find out whether *G. zaddachi* responds to such changes in hydrostatic pressure only. A population of *G. zaddachi* was subjected to a rise in hydrostatic pressure of 0.5 atm. in the current chamber for a short time (15 minutes), by means of a vessel, containing the same water as the chamber, placed on a height of 5 m. The animals were reacting rapidly by swimming actively against the current and close to the water surface (no activity counts were made), but the current chamber could not resist the extra pressure, hence the short duration of this experiment. Another attempt with an increase in hydrostatic pressure of 0.3 atm. gave the same results as mentioned above.

Without technical difficulties the animals were subjected to 8 pressure cycles of tidal periodicity, each with an increase in pressure of 0.15 atm. during 1.5 hours. The activity pattern during 4 of such cycles is shown in fig. 17. The animals do react on a change in pressure of 0.15 atm., but the way they react is quite variable. In one case an activity peak appears during the increase of hydrostatic pressure, in another during or just after the decrease of pressure. The activity under constant conditions after this treatment was on such a low level, that no conclusions can be made about a persistent rhythm.

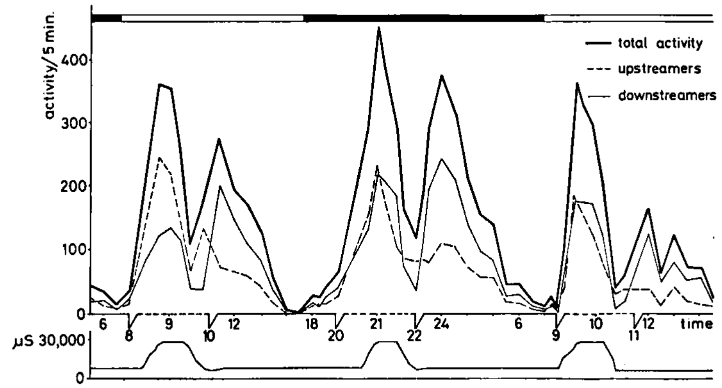

Fig. 15. Influence of changes in salinity on the activity of *G. zaddachi*, showing the characteristic peaks in swimming both during the increase as well as after the decrease in salinity. The 4th, 5th and 6th tidal simulation of the experiment from fig. 12 are shown. For details on experimental conditions and population see fig. 12. Bars indicate light and dark .

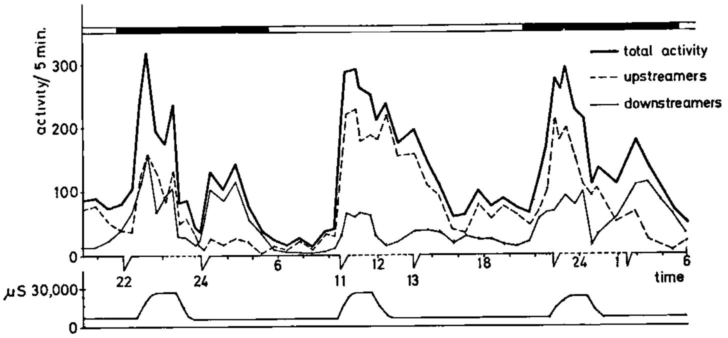

Fig. 16. Influence of light on the rheotactic behaviour of *G. zaddachi*. In the light, upstream swimming dominates during the second activity peak (after the decrease in salinity), whereas in the dark downstream swimming dominates. The 7th, 8th and 9th tidal simulation of the experiment from fig. 13 (with changes in salinity only) are shown. Bars indicate light and dark. For further details see fig. 13.

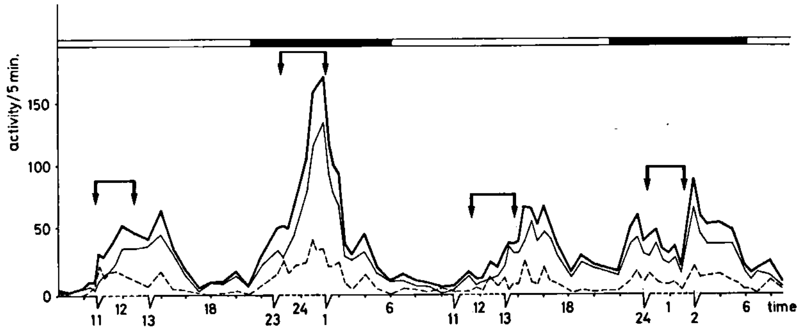

Fig. 17. Activity pattern of *G. zaddachi* during the last four of a series of eight rises in pressure of 0.15 atm. in the period indicated by arrows, at tidal intervals. Further legends as in fig. 16. The animals were caught on 23 March 1979 and 100 specimens were put in the current chamber on 16 April (temperature 10-11 °C, specific conductivity 10^4 μS, V = 3). Survival: 54 animals were still alive on 25 April.

DISCUSSION

In the present paper it is extensively demonstrated that *G. zaddachi* originating from stagnant brackish water reacts on high tide simulations in a current chamber. This corresponds with the conclusion of Girisch & Dennert (1975), based on few experiments only, that the typical behaviour of estuarine *G. zaddachi*, an activity peak at a high tide simulation, is also found in non-tidal populations.

Remarkable is, that the animals react with the same intensity on a high tide simulation (either complete or partial) in the light as they do in the dark. This is in contrast with the field results. Anadromous migration is only found in the hours around nocturnal high tide (Dennert et al., 1969, Girisch et al., 1974). Girisch & Dennert (1975) only did simulations at night time, in view of the fact that upstream as well as downstream migration in the field is confined to the hours of darkness. Consequently, no data were available up to now on the behaviour during simulations in the light.

The master factor triggering the activity at high tide simulations was in our case a rise in salinity. This sole factor causes a reaction resulting in an activity level comparable with that at complete high tide simulations (cf. figs. 15 and 16 with fig. 2).

This is in contrast with the conclusion of Girisch & Dennert (1975), that the master factor, triggering activity at high tide simulations, is in both estuarine and non-tidal populations a change in current velocity and direction. In fig. 8 is shown, that the reaction of our population of non-tidal *G. zaddachi* on such changes is far smaller than that on complete high tide simulations. In the case of a sole change in current velocity and direction the normal nocturnal activity is even dom-

TABLE I

Summary of the activity data in the simulation experiments performed by Girisch & Dennert (1975) with *G. zaddachi*. NS-Canal = North Sea Canal, V = stream velocity, S = salinity, and T = temperature. All activity counts are given as the sum of up- and downstream migrants per minute. The number of animals in the current chamber was about 100 in all experiments.

Fig. no. in Girisch & Dennert, 1975	Last activity count before reduction of stream flow	Highest activity during current reduction & reversal	Net increase of activity	Last activity count before salinity increase	Highest activity during salinity increase	Net increase of activity	Origin of population	Tidal factors included in simulation
Column no. 1	2	3	4	5	6	7	8	9
18	6	38	32	11	42	31	Slack	V, S, T
19	3	13	10	6	11	5	Slack	V, S, T
20	5	32	27	21	46	25	NS-Canal	V, S, T
21	7	22	15	12	26	14	NS-Canal	V, S, T
25	—	—	—	12	24	12	Slack	S
26	4	9	5	7	19	12	Slack	V, S, T
27	—	—	—	25	38	13	NS-Canal	S, T

inating. This is in contrast with the pattern in the series with complete high tide simulations (fig. 8, interrupted line) and with simulations, consisting of changes in salinity only (figs. 12, 13, 15 and 16).

It seems, however, questionable if the results on *G. zaddachi*, presented by Girisch & Dennert (1975) justify their conclusion indeed. A first objection is, that simulations with changes in current only (a step by step decrease of the stream velocity, followed by a standstill and a slow current in the opposite direction, and vice versa) have not been done with *G. zaddachi*, which makes a direct comparison with the sole influence of changes in salinity rather difficult. In table I the results of the simulation experiments with *G. zaddachi* performed by Girisch & Dennert (1975) are summarized. For easier comparison the numbers of upstream and downstream migrants have been summed to obtain the total activity per minute.

There is a clear and rather stable pattern during all simulations with changes in both current and salinity (at the same time combined with a change in temperature). During the reduction in stream velocity swimming activity obviously increases, but drops again during the following period of current reversal and low stream velocity. Sometimes such a decrease is even found during the standstill of the current (Girisch & Dennert, 1975: figs. 20, 21, 26). When seawater is pumped in the chamber, immediately a new peak in swimming activity follows, often even higher than the first peak during the reduction of stream flow (table I: figs. 18, 20, 21 and 26). As seawater is pumped in the chamber in the period with low current velocity, consequently there is often a somewhat higher activity than before the start of the simulation. Therefore it is better to compare the net increase in activity for the first and second peak (table I, columns 4 and 7). In each experiment the net increase in activity is about the same for the two peaks. In some experiments (table I, figs. 19 and 26) a kind of apathy is found and the reaction on both changes in current and salinity is very small. When there is a more overt reaction on the reduction of stream velocity, there is likewise a pronounced reaction on the salinity increase. In the cases that the sole influence of a rise in salinity is examined an increase in activity is found, which is not obviously smaller than that brought about by the changes in current (table I, figs. 25 and 27). In view of this, and of the fact that within the same experiment the changes in current and in salinity provoke increases of swimming activity of the same magnitude, it is possibly better to conclude that both factors clearly influence activity and to avoid a conclusion on the rank of influence.

Another objection is, that almost all simulation experiments performed by Girisch & Dennert (1975) were started at about 22.00 h, just around the normal nocturnal activity peak. This makes it somewhat obscure in how far the observed activity peaks, especially in the experiments with rather small reactions, are due to the tidal stimuli or to the normal nocturnal activity peak. Likewise, in experiments with estuarine *G. zaddachi* the pronounced endogenous tidal and semilunar rhythms, which can occur in such populations (Dieleman, 1977), can influence the response of freshly caught animals to tidal simulations.

The existence of an endogenously controlled tidal activity rhythm in non-tidal *G. zaddachi* is clearly shown in figs. 3, 5 and 6. The behaviour of this population therefore not only resembles that of their estuarine conspecifics with regard to their response on tidal simulations, but also with regard to the presence of a "tidal clock", enabling them to adjust their activity to the rhythmic pattern of the tides.

Our question, which factor is responsable for the entrainment could not be answered completely in so far, that the rhythm could not be induced by one of the tested factors alone. At any rate, the rhythm clearly becomes apparent after a series of complete high tide simulations, even when a change in temperature is left out. Although the rise in salinity is the main factor triggering swimming activity in our experiments, a treatment by changes in salinity at tidal intervals (with additional changes in pressure due to pumping) does not result in a tidal activity rhythm under constant conditions, though there is some indication of recurrent activity at the expected time of high water (figs. 12 and 13).

Tidal cycles with just changes in current flow were clearly insufficient to induce a persistent tidal rhythm, only the normal nocturnal activity pattern being found under constant conditions. Cycles in stream flow were also tested by Rodriguez & Naylor (1972) as to their ability to entrain a persistent tidal activity rhythm in the littoral prawn *Palaemon elegans.* During a ten-day treatment the animals were much more active during the hours with flowing water (6 hours, alternating with 6 hours of stagnant water), but this pattern did not persist under constant conditions.

The small changes in temperature were not tested separately, as Girisch & Dennert (1975) found hardly any effect of such changes on activity. Unlike animals exposed to the air or staying behind in small pools when the tide falls, estuarine *G. zaddachi* rarely undergo temperature shocks of an amplitude higher than 4 °C. Still the most regular persistent rhythm appeared in the experiment of figs. 2 and 3, where a temperature change of 4 °C was incorporated in the tidal simulations, which could mean that temperature changes have a benificial influence on the effectiveness of the simulations in entraining the rhythm.

The rather small reaction of *G. zaddachi* to pressure cycles of 0.15 atm. compared with the very clear response to increases in pressure of 0.3 and 0.5 atm. is possibly due to the fact, that 0.15 atm. is close to the threshold value, though Morgan (1967) still found a response of the crab *Macropipus holsatus* to 0.05 atm., whereas for the amphipod *Synchelidium* even a threshold value of 0.01 atm. has been found (Enright, 1961, 1962). In connexion with the small response to cycles of 0.15 atm. and the absence of a persistent tidal rhythm after treatment by cycles in salinity combined with small changes in pressure of about 0.1 atm., these cycles in pressure due to pumping seem to be a factor of limited importance in our experiments. However, in the estuary, where an increase in pressure of 0.5 atm. during spring tides can occur, this may of course be different.

In conclusion, only complete high tide simulations or simulations with changes in current plus salinity were able to entrain a circatidal activity rhythm in non-tidal *G. zaddachi.* Of course, this does not exclude the possibility that each individual factor, like a change in pressure, salinity, current or temperature, can be an effective synchronizer when applied with a higher amplitude and/or a longer duration, especially when the experiments are performed in constant dark. For instance, Taylor & Naylor (1977) used cycles of 6 hours of normal salinity, alternating with 6 hours of raised salinity, whereas in our experiments the ratio was approximately 11 : 1.5.

Possibly seasonal variations in the response to changes in environmental factors can also play a role (though this seems more likely for estuarine populations). An interesting case in this context is mentioned by Naylor & Atkinson (1972) for the Shore Crab, *Carcinus maenas.*

The fact that a combination of factors is effective, while the individual factors are less so or not, has been more often encountered. Williams & Naylor (1969) found that cycles of immersion at constant temperature were, surprisingly enough, not effective in inducing a tidal rhythm in *Carcinus*. When kept in moist air, the crabs clearly showed a circatidal rhythm after a five-day treatment with cycles in air temperature (6.2 hours at 24 °C / 6.2 hours at 13 °C). Environmentally more realistic cycles of 17 °C / 13 °C were much less effective, but when the 13 °C period was combined with immersion in seawater, the tidal rhythm was much more readily imposed than with the temperature cycle alone.

In the present paper the existence of a circatidal activity rhythm with equal peaks in the light and in the dark is clearly demonstrated, as contrasted with the lunar day (24.8 h) periodicity in upstream migration found in field studies. The inhibiting role of light under field conditions is well known for other amphipods (Jansson & Källander, 1968). It has also been proved experimentally for some species. For instance, the sand-dwelling intertidal amphipod *Bathyporeia pelagica* swims under constant conditions in the laboratory at every expected high water, but in light/dark cycles the peak at daytime high water almost completely disappears. Reversal of the light regime results in swimming activity only at high water during the artificial night (= natural day) (Fincham, 1970b). In the field, swimming is indeed mainly found at night (Fincham, 1970a). Also the Brown Shrimp, *Crangon crangon* shows swimming peaks at every expected high water in constant darkness in the laboratory. When kept in light/dark cycles the peak at daytime high water almost completely disappears, but on the shore push-net catches still suggest a considerable activity during daytime high water, which may be attributed to the light reduction brought about by the increased turbidity at high tide (Al-Adhub & Naylor, 1975).

The data on *G. zaddachi* show just the opposite. In the laboratory, there is no indication at all for inhibition of the tidal activity peak in the light (either as the result of the high tide simulation or as the result of the endogenous tidal rhythm). Thus the strong influence of light, demonstrated by a nocturnal activity pattern before the experiments (fig. 1), completely disappears when the populations are subjected to tidal simulations. So, it is rather puzzling why upstream migration in nature is almost confined to the spring tides in the dark. (Possibly an exception must be made for the spring tides coinciding with sunrise, when Girisch et al., 1974, found enormous amounts of upstreamers in the light. Only a small amount of upstreamers was caught when high water spring tide coincided with sunset, possibly due to the fact that the flood tide started in this case in the light.)

It seemed therefore interesting to do some preliminary observations on the activity of *G. zaddachi* in the estuary. During a first attempt, later abandoned for practical reasons, we tried to catch actively swimming gammarids at high water spring tide in the estuary of the Slack (Pas-de-Calais, France) by means of a net with an aperture of 0.30 × 0.30 m, constructed on 0.15 m high slides to avoid disturbance of the substrate, which was towed by a motorboat. Another attempt by hauling the same net from the bank across the river mouth just downstream of the sluice where the Slack crosses highroad N40 and where the wide estuary is separated from the narrower riverbed (see Dennert et al., 1969, fig. 1), was more succesful. In 5 standard hauls 15 *G. zaddachi* were caught at high water spring tide during daytime and 90 at nocturnal high water spring tide. Possibly this site is not entirely representative of truly estuarine conditions. The sluice doors, poorly functioning during the study of Dennert et al. (1969) and thus permitting the tides to penetrate far upstream, now close completely at high tide. Consequently, there is presently no reversal of the current. Certainly more data on the activity of *G. zaddachi* in the estuary at spring tides in the light and in the dark are necessary, but this could not yet be realized within the scope of the present study.

Under constant conditions in the laboratory *G. zaddachi* from the Slack estuary, sampled just before the highest spring tides, show a well-developed circatidal swimming rhythm, with equal peaks in the light and in the dark (fig. 18), exactly as was found for the induced persistent tidal rhythm in non-tidal *G. zaddachi*. This, in combination with the equal reactions on tidal simulations in the light and in the dark suggests that swimming will also occur in nature at spring tides during daytime. Still, no upstream migration occurs at that time.

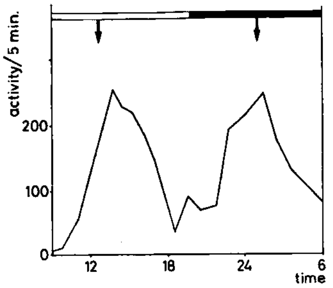

Fig. 18. Activity pattern of *G. zaddachi* from the estuary of the river Slack, sampled at low tide on 27 March 1979, about 6 p.m. Twenty-five animals were put in stagnant water from the sampling site (temperature 9 °C, specific conductivity 10^3 μS), in a small aquarium and observed (after about 12 hours of acclimation) on 28 and 29 March. Fluorescent lamps and a 100 Watt bulb (about 1.5 m above the aquarium) were used during the day and at night counts were made by means of dim red light. Here too, activity was defined as the number of animals crossing a vertical reference line on the aquarium wall per time unit. Bars indicate light and dark; arrows indicate the expected time of high water in the sampling area.

Two alternative explanations for this contradiction between field observations and laboratory results are possible:

(1) No swimming occurs in the field during spring tides at daytime. It is hard to believe that this could be simply the result of the light intensity, because the intensity used in the laboratory (900 lux measured at the water surface) is much higher than at the bottom of the estuary, in view of the high turbidity of the flood waters. For instance, in the Tamar estuary along the British Channel coast, Cooper & Milne (1938) found that the underwater light intensity during flood tide is often reduced to 1 % within 1 or 2 m. Moreover, this alternative would imply that the observations in the laboratory (both the response to simulations and the endogenous peaks) have no predictive value for the behaviour in nature at all, which again is unlikely.

(2) Swimming occurs during spring tides at daytime. This alternative fails to explain why swimming activity at nocturnal spring tides results in upstream migration, whereas the same activity at daytime spring tides does not. Theoretically this could be expected when light would influence the orientation of the gammarids. This, however, is not supported by our results. We did not find that under the influence of light the animals kept a closer contact with the substrate, which could have explained the absence of migratory displacements in the field during daytime spring tides despite of the activity. Also the ratio upstreamers/downstreamers is the same in the light and in the dark (fig. 2). Just in one of the series of high tide simulations, viz. that with changes in salinity only (fig. 16), we found an influence of light on rheotactic behaviour (i.e., during the second activity peak, after the decrease in salinity, animals swimming against the current were dominating at day and those swimming with the current at night).

Possibly the absence of a clear influence of light in the laboratory, both during the simulations and on the expression of the endogenous component, so different from the very strong influence on migration in nature, must be sought in other aspects of light than only its intensity. In our current chambers light always penetrates from all possible directions and due to the very clear water used, there will hardly be a gradient in intensity. Due to the limiting angle, light reaches the animals in nature only from directions within a cone with a half-angle of about 48° (see Weinberg, 1976, for underwater photographs of this phenomenon) and there is an enormous gradient in intensity in the turbid water during high tide. This could result in an orientation towards the substrate at high water spring tide during daytime, and such an increased contact with the substrate could in turn result in a strong reduction of migratory displacements compared with high water in the dark, when the animals swim higher up in the water.

More detailed information on the emergence and swimming activity during daytime spring tide is therefore urgently needed for a better understanding of the way in which light influences the effect of tidal factors like changes in current, salinity, temperature and pressure as well as the expression of the endogenous tidal activity rhythm. At first, net catches with the method described above will be useful. Probably underwater video records could also be profitable.

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REFERENCES

Aarons, D. & M. Reagan, 1977. SPSS Spectral, Spectral analysis of time series. Vogelback Computing Center, Northwestern University, Everton, Ill., Manual **434**: 1-24.

Al-Adhub, A. H. Y. & E. Naylor, 1975. Emergence rhythms and tidal migrations in the Brown Shrimp Crangon crangon (L.). J. mar. biol. Ass. U.K., **55**: 801-810.

Arnold, G. P., 1974. Rheotropism in fishes. Biol. Rev., **49** (4): 515-576.

Cooper, L. H. N. & A. Milne, 1938. The ecology of the Tamar estuary. II. Underwater illumination. J. mar. biol. Ass. U.K., **22**: 509-527.

Dennert, H. G., A. L. Dennert, P. Kant, S. Pinkster & J. H. Stock, 1969. Upstream and downstream migrations in relation to the reproductive cycle and to environmental factors in the amphipod, Gammarus zaddachi. Bijdr. Dierk., **39**: 11-43.

Dennert, H. G. & M. J. van Maren, 1974. Further observations on the migration of Gammarus zaddachi Sexton (Crustacea, Amphipoda) in a French stream. Bull. zool. Mus. Univ. Amsterdam, **3** (20): 157-167.

Dieleman, J., 1977. Circatidal activity rhythms and the annual migration cycle in an estuarine population of Gammarus zaddachi Sexton, 1912. Crustaceana, Suppl. **4**: 81-87.

Enright, J. T., 1961. Pressure sensitivity of an amphipod. Science, N.Y., **133**: 758-760.

——, 1962. Responses of an amphipod to pressure changes. Comp. Biochem. Physiol., **7**: 131-145.

Fincham, A. A., 1970a. Amphipods in the surf plankton. J. mar. biol. Ass. U.K., **50**: 177-198.

——, 1970b. Rhythmic behaviour of the intertidal amphipod, Bathyporeia pelagica. J. mar. biol. Ass. U.K., **50**: 1057-1068.

Girisch, H. B. & H. G. Dennert, 1975. Simulation experiments on the migration of Gammarus zaddachi and Gammarus chevreuxi. Bijdr. Dierk., **45** (1): 20-38.

Girisch, H. B., J. C. Dieleman, G. W. Petersen & S. Pinkster, 1974. The migration of two sympatric gammarid species in a French estuary. Bijdr. Dierk., **44** (2): 239-273.

Jansson, B. O. & C. Källander, 1968. On the diurnal activity of some littoral pericarid crustaceans in the Baltic Sea. J. exp. mar. Biol. Ecol., **2**: 24-36.

Jones, D. A. & E. Naylor, 1970. The swimming rhythm of the sand beach isopod Eurydice pulchra. J. exp. mar. Biol. Ecol., **4**: 188-199.

Klapow, L. A., 1972. Natural and artificial rephasing of a tidal rhythm. J. comp. Physiol., **79**: 233-258.

Morgan, E., 1965. The activity rhythm of the amphipod Corophium volutator (Pallas) and its possible relationship to changes in the hydrostatic pressure associated with the tides. J. anim. Ecol., **34**: 731-746.

——, 1967. The pressure sense of the swimming crab Macropipus holsatus (Fabricius), and its possible role in the migration of this species. Crustaceana, **13**: 275-280.

Morgan, E., A. Nelson-Smith & E. Knight-Jones, 1964. Responses of Nymphon gracile (Pycnogonida) to pressure cycles of tidal frequency. J. exp. Biol., **41**: 825-836.

Müller, K., 1963. Diurnal rhythm in organic drift of Gammarus pulex. Nature, Lond., **198**: 806-807.

Naylor, E. & R. J. A. Atkinson, 1972. Pressure and the rhythmic behaviour of inshore marine animals. Symp. Soc. exp. Biol., **26**: 395-415.

Palmer, J. D., 1973. Tidal rhythms: the clock control of the rhythmic physiology of marine organisms. Biol. Rev., **48**: 377-418.

Rodriguez, G. & E. Naylor, 1972. Behavioural rhythms in littoral prawns. J. mar. biol. Ass. U.K., **52**: 81-91.

Taylor, A. C. & E. Naylor, 1977. Entrainment of the locomotor rhythm of Carcinus by cycles of salinity change. J. mar. biol. Ass. U.K., **57**: 273-277.

Weinberg, S., 1976. Submarine daylight and ecology. Mar. Biol., **37**: 291-304.

Williams, B. G. & E. Naylor, 1969. Synchronization of the locomotor tidal rhythm of Carcinus. J. exp. Biol., **51**: 715-725.

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