

PERISTALSIS IN THE MONOPOLYPID STAGE OF THE SEA-PEN, *CAVERNULARIA OBESA VALENCIENNES*¹⁾

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With Text-figures 1-9

Abstract

Peristalsis in the monopolyrid stage of the sea-pen, *Cavernularia obesa*, was studied experimentally as to the change in light, temperature and pH of the sea water.

(1) The monopolyrid of this sea-pen shows two types of peristalses: downward and upward peristalses.

(2) Downward peristalsis occurs more frequently in the light phase, while upward peristalsis is mostly limited to the dark phase.

(3) Downward peristalsis is induced by sudden illumination at night, with the frequency depending on the absolute light intensity as well as the variation in light intensity.

(4) The frequency of downward peristalsis is seemingly temperature-dependent.

(5) The frequency of downward peristalsis is more or less decreased in the sea water of lowered pH, but increased in the sea water of raised pH, while the frequency of upward peristalsis seems to be rather insensitive to pH change.

(6) On the basis of these results, peristalses of the monopolyrid of this sea-pen are discussed in comparison with those of the adult colonies.

Introduction

The peristalsis has been known in adult colonies of sea-pens such as *Renilla* (Parker, 1920), *Pteroides* (Brafield, 1969), *Veretillum* (Ceccatty and Buisson, 1965; Buisson, 1971) and *Cavernularia*, but only briefly referred to in their juvenile bodies in a few papers dealing with *Renilla* (Wilson, 1883) and *Cavernularia* (Mori and Tanase, 1973).

The present paper concerns the peristalsis in the monopolyrid stage of the sea-pen, *Cavernularia*, to see its occurrences, effects of light, temperature and pH on them and the diurnal alternation of direction in this movement.

Miyajima (1897, 1900) was the first to observe the peristalsis in adult colonies of *Cavernularia obesa*, in which the animal laid on the sand and illuminated on the peduncle showed the peristalsis travelling from the illuminated part (peduncle) to the rachis and this movement carried the colony in the direction toward the rachis, while the colony moved in the opposite direction by peristalsis travelling from the rachis to the peduncle, that was induced by illumination on the rachis. Honjo (1940)

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described some types of peristalses in the colonies of *Cavernularia* which were kept laid in the glass vessel. Mori's observation (1943) which was made on the colonies set in the sand in the natural posture revealed that the peristalsis travelled from the peduncle to the rachis in the expanding phase of the colony but in the opposite direction in the course of contraction. Such a relation between the direction of peristalsis and the expansion-contraction behavior was also observed in another species of sea-pen, *Veretillum cynomorium* (Ceccatty and Buisson, 1965).

In *Cavernularia*, the peristalsis was observed in the planula older than 2 days old and the monopolyrid (Mori and Tanase, 1973).

Materials and Methods

The experiments were made in 1971 to 1973 at the Seto Marine Biological Laboratory. Materials used were juvenile bodies of the monopolyrid stage of *Cavernularia obesa* Valenciennes, which had been developed from the eggs spawned in the aquarium in October to November. Mother colonies were collected from Hatakejima Island just before the spawning season. Spawned eggs were kept in glass bowls, 6 cm in diameter and 3 cm high, and monopolyrid juveniles developed from these eggs settled on the bottom of bowls. The number of juveniles in each bowl never exceeded five. The sea water in these bowls was exchanged during the development and experiments every one to five days, and the water temperature was kept the same as in the running sea water of the laboratory, in which the bowls were kept immersed. The bowls were exposed to the natural daylight or to an artificial illumination. No feeding was made through the development and experiments, as the suitable food of juveniles was unknown, so that they did not survive more than 30 days.

Results

I. *Peristalsis*.

The present author has ever heard from Mr. Tanase that two types of peristalses were observable in the monopolyrid stage of the sea-pen, *Cavernularia*, the peristalsis proceeding from the rachis-like part to the tip of peduncle (the downward peristalsis) and that in the opposite direction from the tip of peduncle to the rachis-like part (the upward peristalsis). Firstly, this was confirmed by the present author himself.

I-1. Downward peristalsis.

The downward peristalsis of a higher frequency was observed in the light phase of an artificial light-dark cycle (see IV. Diurnal alternation of direction in peristalsis) and induced by illumination at night (see II. Effect of light).

The peristalsis begins on a polyp laid still on the bottom of the bowl with the sudden swing of the probable precursory rachis inclusive of the polyp body (Fig. 1a). Soon after, a contraction wave appears just above the buds of the secondary polyps (Fig. 1b-c) and slowly travels downward (Fig. 1d-g) till it disappears at the tip of

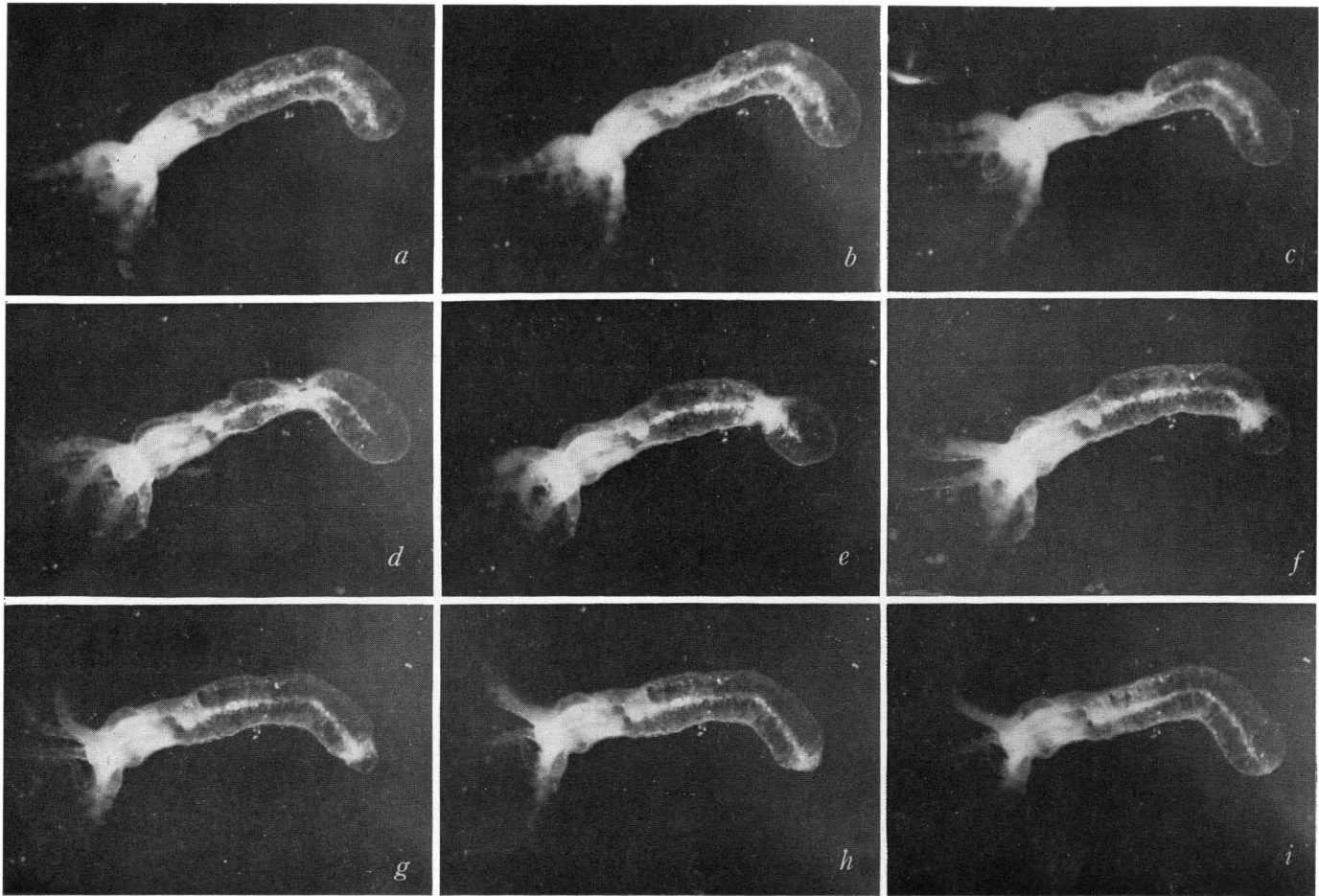


Fig. 1. The downward peristalsis induced at night by illumination on a 2.8 mm long monopolypid body. The wholly relaxed body looks transparent, while the contracted part somewhat opaque.

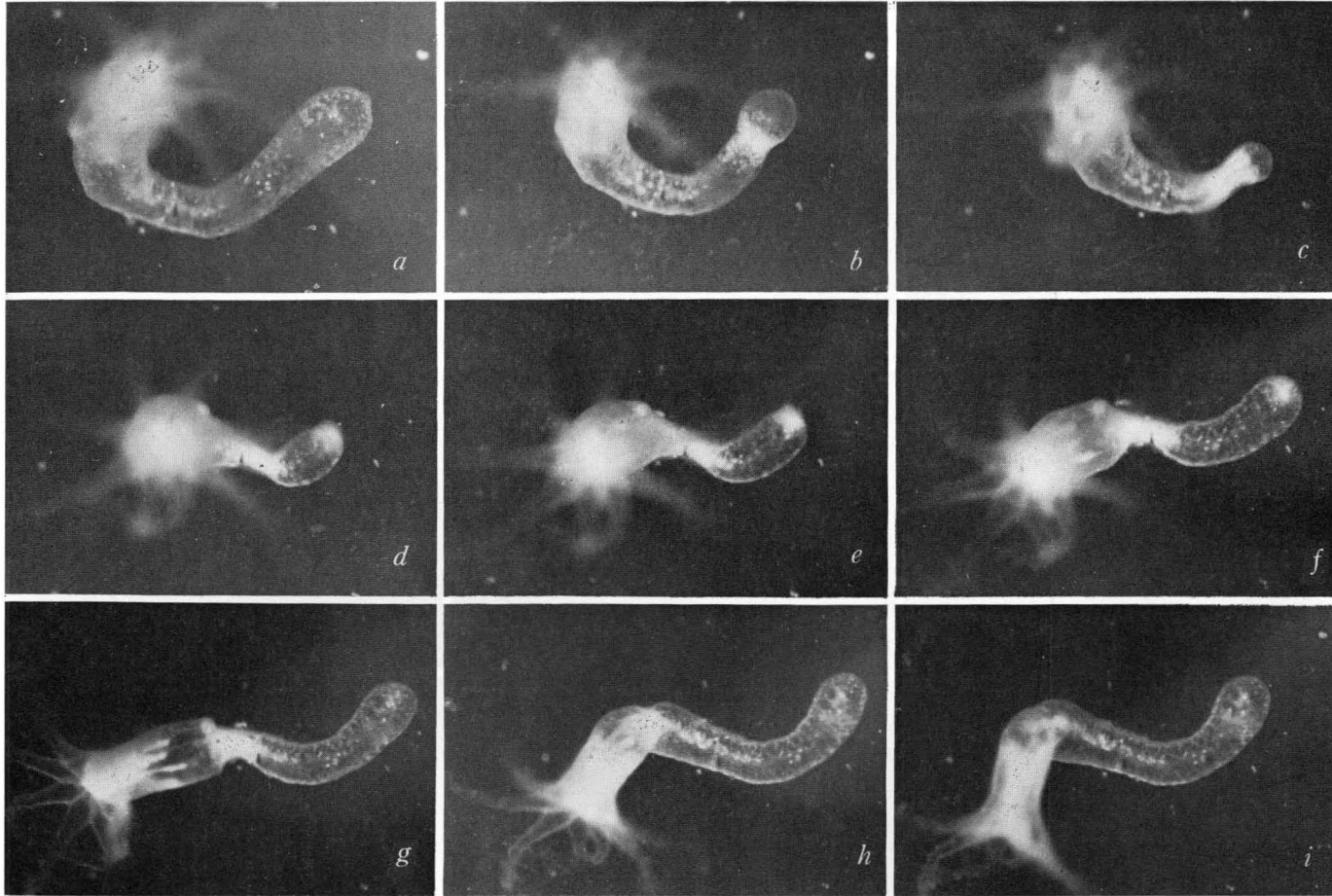


Fig. 2. The spontaneous upward peristalsis maintained on a 3.0 mm long monopolyrid body in the dim light condition without any inducement. See the note in Fig. 1.

the peduncle (Fig. 1h-i).

The whole process needs 15 to 60, but usually 20 to 30 seconds. As the peduncle is 2 to 4 mm long, the travelling rate of the contraction wave may be 2 to 16 mm/min. The rate is seemingly affected by environmental factors such as the light intensity and temperature, though this has not yet been closely investigated. Intervals of successive contraction waves are 15 seconds or more, but rarely another contraction wave may appear before a wave completely disappears and thus two waves are simultaneously observed on a single juvenile body.

I-2. Upward peristalsis.

The upward peristalsis was observed in the dark phase of an artificial light-dark cycle; this phase did not mean the real darkness but a dim light condition to allow observations (see IV. Diurnal alternation of direction in peristalsis).

As in the downward peristalsis, shaking of the precursory rachis and the polyp body precedes the appearance of a contraction wave beginning *near the tip* of the peduncle in the upward peristalsis, too (Fig. 2b). As seen in Figure 2, a contraction wave generated near the tip of the peduncle (Fig. 2b) is splitted into two (Fig. 2c), the minor one travelling toward the tip of the peduncle and disappearing there (Fig. 2f-g), while the major one travelling to the precursory rachis (Fig. 2d-f) and disappearing just above the buds of the secondary polyps (Fig. 2g-i). Thus, the wave travelling upward is always much more conspicuous than that travelling downward, so that the latter may sometimes become quite obscure. However, when a contraction wave appears first in *the middle part* of the peduncle, the splitted two waves respectively travelling upward and downward are both very clear.

The time necessary for the whole process of the upward peristalsis is 20 to 60 seconds and the travelling velocity of the wave is estimated to be 2 to 12 mm/min. The appearance of another contraction wave before the complete disappearance of the preceding one, that is observable in the downward peristalsis, has never been observed in the upward peristalsis. The upward peristalsis once induced will never be stopped or disappear till the whole process is completed, even under the illumination enough to take photographs.

II. *Effect of Light.*

As the preliminary experiments showed that the downward peristalsis was induced at night by illumination, the effect of the light intensity on the frequency of this movement was investigated by exposing a monopolypid body to the bright and low light or keeping it under constant illumination.

II-1. Bright light.

A monopolypid body (P-11) of 8 days old was experimented with various light changes at night, from 21:00, 30th to 0:40, 31st of October, 1971. Prior to the experiments, it had been kept since dusk under illumination of 400 lux. The light source was a tungsten lamp and its intensity was controlled by changing the voltage and measured at the same distance from the source as that between it and bowl containing the monopolypid body. The water temperature in the bowl was 19.8

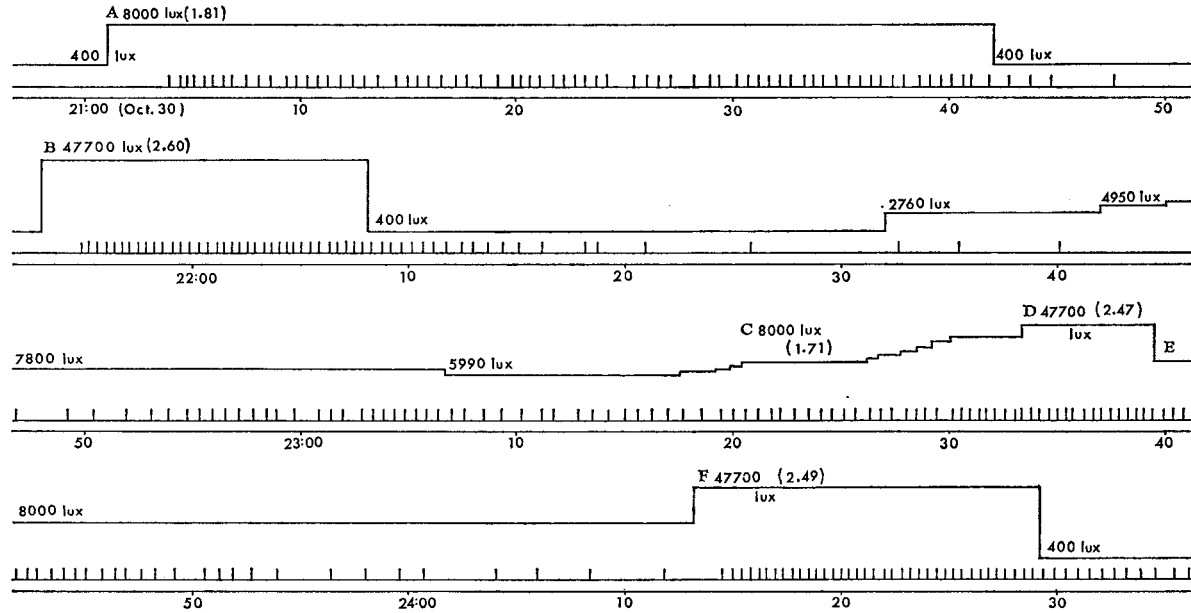


Fig. 3. Effect of bright light on the frequency of downward peristalsis in a monoplypid body (P-11) kept at 400 lux prior to experiments. (A) Exposure to the sudden illumination of 8000 lux, (B) of 47700 lux. (C) Exposure to the gradual rise of light intensity to 8000 lux and (D) further to 47700 lux. (E) Exposure to a drop of light intensity from 47700 to 8000 lux. (F) The last exposure to a rise of light intensity from 8000 to 47700 lux. The frequencies at respective light intensities marked with A-F are shown in parentheses.

$\pm 0.1^{\circ}\text{C}$. Respective peristalses observed are recorded in Fig. 3 by showing, with a short vertical bar, the time of disappearance of waves at the tip of peduncle.

When the monopolypid body under the illumination of 400 lux was exposed suddenly to a bright light of 8000 lux (A), the downward peristalsis was induced at a lag of 175 seconds, in that time the tentacles showed some movements but the peduncle without any responses. A contraction wave once appeared continued one after another during the exposure to the bright light. The interval of successive contraction waves was not always constant but fluctuated between 20 and 70 seconds. Then the frequency of the contraction wave at 8000 lux was estimated to be 1.81/min on an average, that is, the average interval of successive waves was 33.1 seconds. After 41 minutes of exposure to 8000 lux, the light intensity was lowered again to 400 lux and then the downward peristalsis became gradually less prominent and finally disappeared.

After a rest for 11 minutes at 400 lux, the monopolypid body was exposed suddenly to 47700 lux (B); this induced the peristalsis of a higher frequency at a lag of 115 seconds. This time, the average was 2.60/min. The light intensity was dropped to 400 lux again, and then the animal became gradually inactive till the peristalsis disappeared at last.

After a pause for about 24 minutes at 400 lux, the monopolypid was exposed to a gradual rise of the light intensity from 400 lux to 8000 lux, which was achieved in about 48 minutes though a slight drop of the intensity was inserted as seen in the figure (C). The downward peristalsis was induced by this at a frequency of 1.71/min, slightly lower than in the case of sudden increase to the same intensity (A).

Continuously, the light intensity was raised gradually to 47700 lux in 13 minutes (D). The frequency rose to 2.47/min, but this was a little lower than in the case of sudden increase to the same intensity (B), too.

After the illumination at the highest intensity for 6 minutes, the intensity was suddenly dropped to 8000 lux (E), then the frequency gradually decreased but much below the level maintained at the first experiment at 8000 lux (A), in 34 minutes. These (C-E) suggest that the change of the light intensity is also an important factor affecting the frequency of peristalsis as well as the intensity itself.

Lastly, the intensity was raised from 8000 lux to 47700 lux (F) and this recovered a high frequency of 2.49/min, that was not so high as in the raise from 400 lux to 47700 lux (B).

From these results, the following conclusions are deducible, if the fatigue effect of the animal body is insignificant: (1) The higher the light intensity is, the higher is the frequency of the downward peristalsis (A and B). (2) A sudden rise of the intensity is a little more stimulative than a gradual rise (A vs. C; B vs. D). (3) The frequency of the downward peristalsis is affected not only by the absolute light intensity but also by increase or decrease of the intensity, inclusive of the changed range and changing rate (A and E; B and F).

II-2. Light of lower intensities.

It must be unnatural that the sea-pen, *Cavernularia*, living on the bottom of

shallows is exposed to the light of 47700 lux as in the previous experiments. Thus, it was planned to examine the effect of the light of lower intensities.

A monopolypid body (P-21) of 11 days old having been kept in the natural day light and then in complete darkness at night was illuminated with an incandescent lamp (20:00 to 22:00). The water temperature was 19.1–19.4°C during the experiments. The light intensity was raised step by step from 80 to 213, 500 and further to 1400 and 3400 lux, and lastly dropped to 500 and then to 213 lux.

The frequency of the downward peristalsis at respective light intensities is shown in Table 1. When the monopolypid was exposed to 80 lux, a downward peristalsis with a frequency of 1.20/min was induced and the frequency rose to 1.45/min at 213 lux. It was strange, but the frequency decreased to 1.08/min at 500 lux; this was lower than those observed at previous lower intensities. The frequencies then increased to 1.30 and 1.41/min at 1400 and 3400 lux, respectively. At dropping of the intensity to 500 and 213 lux, the frequency decreased to 0.91 and 0.87/min, respectively; these were clearly lower than those seen at the same intensities in the raising process.

Table 1. Effect of the light of lower intensities. Frequencies of downward peristalsis measured under the illumination of 80 to 3400 lux. The light intensity was changed in the order from the top to bottom of the left column in the table. The figure in parentheses in the right column indicates the number of peristalses observed.

Light Intensity (lux)	Frequency (per minute)
80 (17 min)	1.20±0.26 (13)
↓	
213 (8 min)	1.45±0.28 (11)
↓	
500 (8 min)	1.08±0.16 (9)
↓	
1400 (8 min)	1.30±0.24 (10)
↓	
3400 (11 min)	1.41±0.14 (14)
↓	
500 (10 min)	0.91±0.31 (9)
↓	
213 (3 min)	0.87±0.12 (2)

The following conclusions may be allowed on these results; (1) There was seen a tendency under the illumination with the light of the range from 80 to 3400 lux, too, that a downward peristalsis of higher frequency was induced by the light of higher intensities, though some irregularity was observed. (2) The light of the same intensity (213 and 500 lux) was differently effective, more stimulative when the light increased to that intensity but less so when the light decreased to that intensity. Here, the consideration on acclimation was put aside.

II-3. Constant light.

In the previous experiments with the light of lower intensities, a regular relation was not observed between the light intensity and the peristaltic frequency. Seemingly this is attributable to such factors as the fatigue of the peristaltic movement and the acclimation to the light under a long time of exposure to the light; these will make the animal become insensitive to or misjudge the intensity difference less than certain magnitude. Then, basically to see how the peristaltic frequency fluctuates or declines under a constant illumination, an experiment was made as follows.

A monopolyid body (P-22) of 12 days old reared in the natural day light and at $25.0 \pm 0.1^\circ\text{C}$ was illuminated at night for 30 minutes (19.13-19.43) at 1700 lux of an incandescent lamp.

The downward peristalsis observed on this monopolyid is shown in Fig. 4. The peristalsis was induced at a lag of 240 seconds, with the frequency relatively high at the beginning, soon decreased, and thereafter fluctuating between 1.5 and 3.0/min.

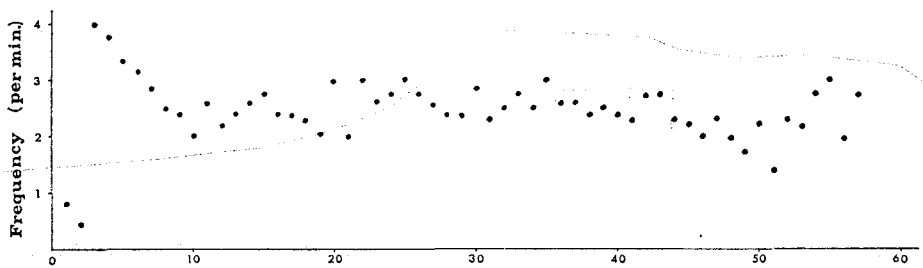


Fig. 4. Effect of constant light observed on a monopolyid body (P-22) exposed to a constant illumination of 1700 lux at 25°C . Ordinate: frequency expressed by reciprocal of the interval between consecutive peristalses. Abscissa: the number of successive peristalses.

From the analysis with the regression line, the frequency was found to decrease by 0.5/min through 100 peristaltic movements.

This seems to show generally that the peristaltic frequency at a certain light intensity can never be always constant for a long time but gradually decreases slightly with time, fluctuating in some range. This will be influential when the change of the light intensity. Minor change or change in the range of lower light intensities might be non-stimulative or fail to induce regular reactions. The above-mentioned phenomena were also observable on the monopolyid bodies kept in an artificial light-dark cycle with the continuously illuminated phase for 12 hours (see chapter IV. Diurnal alternation of direction in the peristalsis).

III. Effect of Temperature.

The expansion-contraction rhythm in the sea-pen is known to be temperature independent, the temperature-coefficient being 1.0 (Imafuku, 1973). On the other hand, the rhythms of higher frequency are generally said to be temperature dependent. Then, the effect of temperature on the frequency of the downward peristalsis was

checked.

III-1. On the peristalsis induced by bright light.

The downward peristalsis was induced by a bright light of 47700 lux at different temperatures and the frequencies were recorded. The experiment was made at night from 22:00 to 1:00 the following morning with the monoplypid (P-11) that had ever been used in the experiments for the bright light and then was 9 days old.

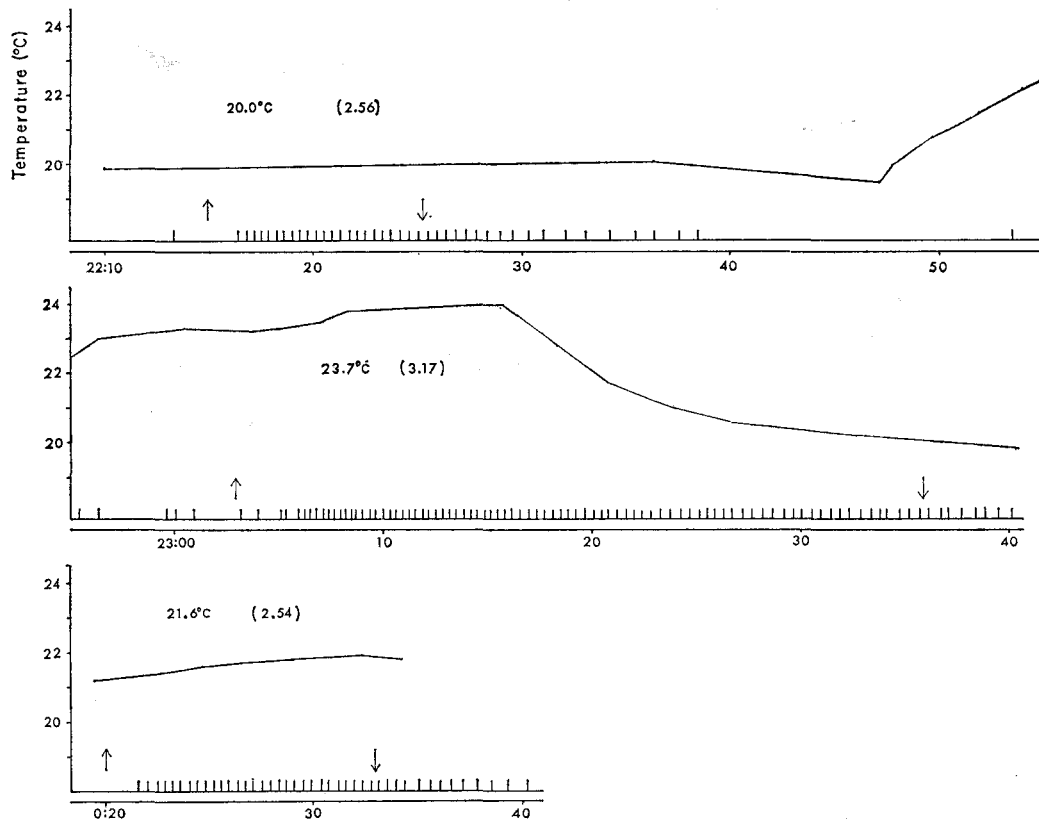


Fig. 5. Effect of temperature on the frequency of downward peristalsis of a monoplypid body (P-11), induced by bright light of 47700 lux at 20.0, 23.7 and 21.6°C. The frequency at each temperature is shown in parentheses. Upward and downward arrows show respectively switch on and off for the bright light.

The monoplypid kept at 20°C and at 400 lux was exposed to the bright light, this induced a peristalsis with a frequency of 2.56/min (Fig. 5). After 10 minutes the light was returned to 400 lux, and after 22 more minutes the ambient temperature was gradually raised. This temperature rise induced some peristaltic movements, 6 minutes after the start, and the animal was exposed again to the bright light when the temperature attained to 23.7°C. This induced the peristalsis with a frequency of 3.17/min. Then, the temperature was gradually dropped for 20 minutes under the

same illumination; this seemed to cause a slight decrease of the frequency. The light intensity was dropped to 400 lux and the temperature was set at 21.6°C. This condition continued for 30 minutes, then the peristalsis was induced once more by the bright light. This time the frequency was nearly the same as that observed first at 20°C.

The highest frequency (3.17/min) was observed at the highest temperature (23.7°C), while the frequency was almost the same at 20°C and 21.6°C. The latter may be attributable to the acclimation of the animal to the light under often exposures to bright light, the smallness of temperature difference, or to the insignificance of the temperature effect in that lower temperature range (20–21.6°C). Anyhow, from this experiment, it may be safely concluded that the downward peristalsis induced by the bright light is temperature dependent at least in the temperature range from 21.6 to 23.7°C.

III-2. On the peristalsis induced by light of lower intensities.

In this experiment, three monopolypid bodies (P-21, P-23, P-24, all 13–14 days old) were exposed respectively to the light of 1300, 1800 and 2600 lux of an incandescent lamp at night at different temperatures. The experiments were made in two days, raising of temperature on the first and lowering on the second day, and respective monopolypids were kept quietly in darkness for 60 minutes prior to the exposure to the light for 20 minutes for respective observations. In the first-day experiment, the temperature was raised from 18.1°C to 21.8°C and finally to 24.9°C in the step method, so that the temperature was raised at the end of each observation to the degree for the next observation. After the first-day experiment was closed, the temperature was set at 26°C for the second-day experiment, in which the temperature was dropped from 26.2°C to 21.1°C and finally to 18.8°C, and every treatment was done as in the first-day experiment.

The frequency measured on each animal at each temperature is shown in Fig. 6. In raising the temperature (solid lines), monopolypids P-21 and P-23 showed relatively high peristaltic frequencies (0.8–1.5/min), while the monopolypid P-24 a very low frequency (0.1–0.3/min). The frequency decreased in P-21 and P-23 but remained the same in P-24, when the temperature was raised from 18.1 to 21.8°C; however it slightly increased in all monopolypids when the temperature was raised from 21.8°C to 24.9°C. In lowering the temperature (dotted lines), the frequency decreased clearly with temperature in the three monopolypids. However, in this experiment, the range of frequency was much higher at all temperatures (1.5–2.7/min) than in the experiment on the first day (0.1–1.5/min).

From these results the following conclusions may be deducible: (1) The frequency of the peristalsis induced by light is higher in the animals having been kept at higher temperatures for a long time, as the frequency in the monopolypids kept at ca. 18°C for a long time before the experiment on the first day was always lower than that in the same animals kept at 26°C for about a day till the experiment on the next day. (2) As the frequency drop with the temperature fall is slightly sharper than the change with the temperature rise, it is suggested that the peristalsis is temperature-dependent.

Further, the frequency tends to decrease as the illumination is repeated, and this might make the temperature effect obscure in some serial experiments.

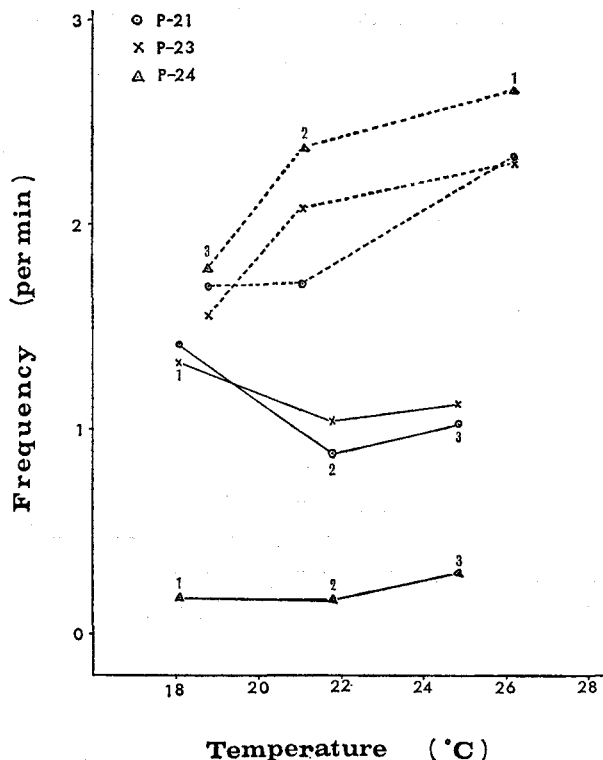


Fig. 6. Effect of temperature on the frequency of downward peristalsis induced by light of lower intensities. Three monopolyptids P-21 (○), P-23 (×) and P-24 (△) were illuminated with the light of 1300, 1800 and 2600 lux, respectively, at 18.1, 21.8 and 24.9°C, raising the temperature in this order on the first day (solid lines), but at 26.2, 21.1 and 18.8°C, dropping the temperature on the next day (dotted lines).

IV. Diurnal Alternation of Direction in Peristalsis.

Having heard from Mr. Tanase that the contraction wave does not always travel in one way, but travels sometimes downward, while upward at other time, the travelling direction of the peristaltic wave was checked in respective phases of an artificial light-dark cycle.

IV-1. In monopolyptid bodies of 14-15 days old.

Three monopolyptids (P-25, P-26 and P-27) developed from the eggs spawned on October 30 had been kept since November 11 under an artificial light-dark cycle of 12 hour light (2000 lux) and 12 hour dark phase of dim light (30 lux) and at 19.0 to 20.0°C, and two to three days later observations were made respectively at the end of the dark phase (D-3), beginning (L-1), middle (L-2), and the end (L-3) of the light phase, beginning (D-1) and the middle (D-2) of the dark phase. Each obser-

vation was made in 30 minutes, but in 29 minutes at L-3. The time of disappearance of respective contraction waves was recorded as seen in Fig. 7.

At the end of the dark phase (D-3), the three monopolypids showed the downward

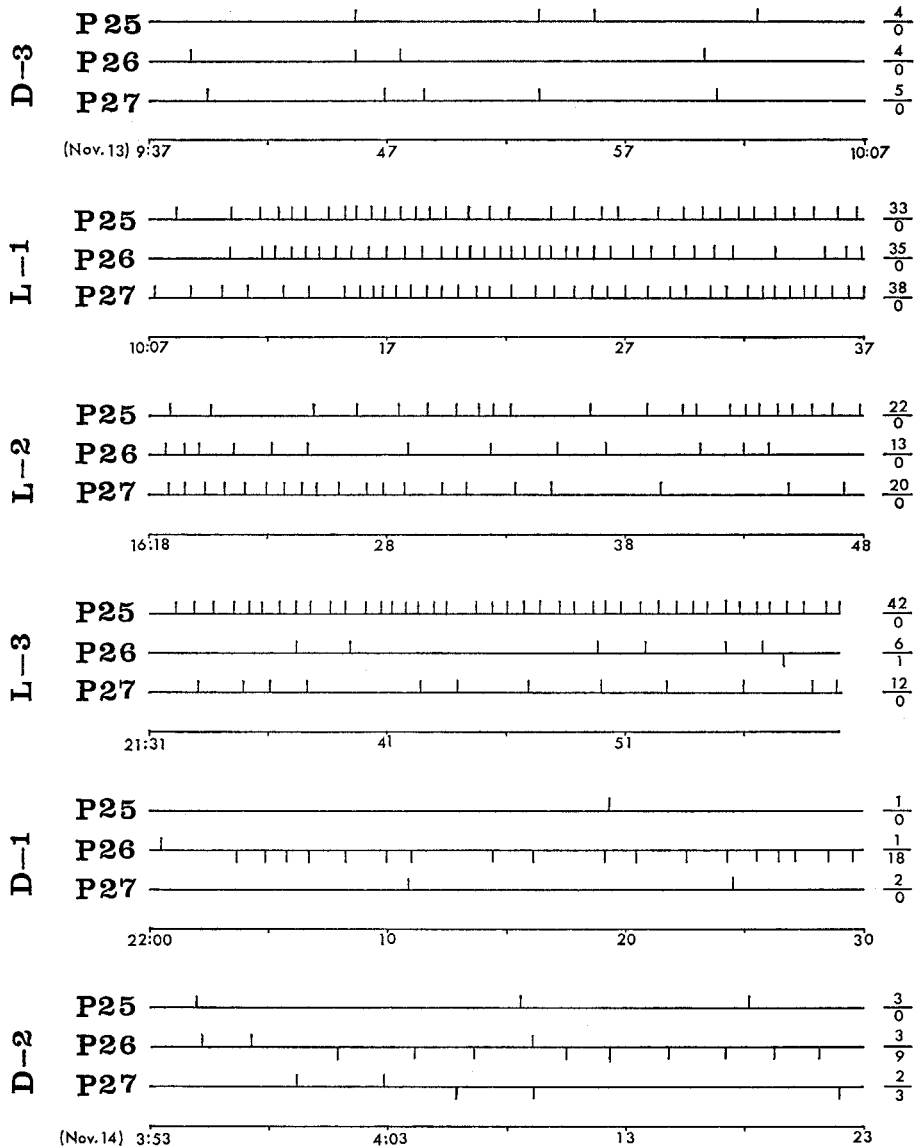


Fig. 7. Diurnal alternation of direction in peristalsis. Three monopolypid bodies (P-25, P-26 and P-27) kept under the cycle of the light phase for 12 hour and the dark phase of dim light for the same hours were observed at the beginning (L-1), middle (L-2) and end (L-3) of light phase, and the beginning (D-1), middle (D-2) and end (D-3) of dark phase for 29 (L-3) to 30 minutes. Downward and upward peristalses are marked with short bars respectively above and below the horizontal line. The number of peristalses per 30 minutes is given on the right side of the figure.

peristalsis of a low frequency. At the beginning of the light phase (L-1), the downward peristalsis of a high frequency was observed in all monopolytoids. In the middle of the light phase (L-2), the frequency of the observed downward peristalsis was not so high as in L-1. At the end of the light phase (L-3), the monopolytoid P-25 showed a distinctly higher frequency while P-26 and P-27 maintained the downward peristalsis of low frequencies; the monopolytoid P-26 showed even an upward peristalsis at the end of this observation. At the beginning of the dark phase (D-1), the monopolytoid P-25 showed only once the downward peristalsis and P-27 merely two downward peristalses, while only the monopolytoid P-26 showed 18 upward peristalses after a downward peristalsis at the beginning. In the middle of the dark phase (D-2), the monopolytoid P-25 exhibited three downward peristalses of a low frequency, while P-26 and P-27 performed upward peristalses intervened by some downward movements.

Results of these observations may be concluded as follows. (1) The downward peristalsis maintained higher frequencies in the light phase. (2) The upward peristalsis was mainly limited to the dark phase. Thus, the downward peristalsis is thought to be related to the light phase, while the upward peristalsis to the dark phase.

IV-2. In monopolytoid bodies of 9-16 days old.

In order to confirm the conclusions given above five more monopolytoids (P-31, P-32, P-33, P-34 and P-35) of 9 to 16 days old were experimented with similarly. This time, these animals had been kept since October 21 under the alternation of the light phase (1000-2000 lux) for 12 hours (10:00-22:00) and the dark phase of dim light (20-50 lux) for 12 hours, and at 22.5 to 24.5°C. But, the light and dark phases of the cycle were reversed on October 28, with the light phase from 22:00 to 10:00. Monopolytoids P-31, P-32 and P-33 were observed on October 25 and 26, and on October 30 to November 1. Therefore, their last observations were made after the phase reversal that was done when they were 12 days old. While the monopolytoids P-34 and P-35 were observed on November 2 to 4 after the phase reversal that was done when they were 9 days old. The observations for 30 minutes were made respectively at the end of the dark phase (D-3), the beginning (L-1) and end (L-3) of the light phase and at the beginning of the dark phase (D-1).

The results of observations shown in Table 2 may be summerized as (1) the upward peristalsis was limited to the dark phase except for only one movement of P-31 in L-1 on October 30, and (2) the frequency of the downward peristalsis was generally higher in the light phase than in the dark phase, though these were a few exceptions. After all, it was proved that the alternation of direction in the peristalsis was under the control of the light-dark cycle, because the alternation of the peristaltic direction was easily reversed by reversing the light and dark phases in the cycle.

IV-3. Relation between the peristaltic direction and the state of tentacles.

It is reported that the state of tentacles of the monopolytoid of the present sea-pen shows a diurnal change, closing in the daytime and opening at night (Mori and Tanase, 1973). To confirm this, the state of tentacles was observed in relation to the peristalsis.

Table 2. The peristaltic frequencies under the light-dark cycle, observed for 30 minutes at the beginning (L-1) and end (L-3) of light phase, and at the beginning (D-1) and end (D-3) of dark phase. u; upward peristalsis. d: downward peristalsis.

	Date	Age (days)	D-3		L-1		L-3		D-1	
			d	u	d	u	d	u	d	u
P31	10/25	9	0	4	30	0	16	0	0	8
	26	10	0	12	24	0	7	0	0	2
	30	14	5	0	36	1				
	31	15	4	1	29	0				
	11/1	16	3	2	28	0	4	0	1	5
P32	10/25	9	7	0	34	0	30	0	22	0
	26	10	9	0	24	0	25	0	25	0
	30	14	6	1	20	0				
	31	15	7	8	29	0				
	11/1	16	8	3	15	0	11	0	1	8
P33	10/25	9	6	3	42	0	10	0	0	5
	26	10	3	4	33	0	16	0	9	0
	30	14	4	0	28	0				
	31	15	0	13	12	0				
	11/1	16	5	2	16	0	3	0	0	10
P34	11/2	14	4	7	40	0				
	3	15	0	10	44	0	22	0	7	0
	4	16					36	0	11	2
P35	11/2	14	5	1	43	0				
	3	15	13	2	39	0	16	0	5	3
	4	16					10	0	2	24

The same monopolypids used in the previous experiment, but then 19 to 23 days old were used in this experiment. The state of tentacles was recorded as *o* (open), *c* (closed) and *i* (intermediate state), corresponding respectively to forms 1, 5 and 3 in the description of Mori and Tanase (1973). Sometimes, tentacles of some monopolypid were found in the transition phase between expansion and contraction at the observation time and then recorded by seeming dominant state. The light-dark cycle followed the last pattern in the previous experiment, with the light phase from 22:00 to 10:00. The water temperature was maintained at 21.0 to 21.9°C. Observation were made arbitrarily, each time for 5 to 10 minutes.

The results of observations are shown in Fig. 8. Monopolypids P-34 and P-35 showed clearly correlated diurnal rhythms in the peristalsis and tentacle state. However, in the monopolypid P-33 the peristaltic rhythm was observed but the tentacle was kept closed throughout the observation, and further in the monopolypid P-32, all peristalses travelled downward and tentacles were kept closed and in the monopolypid P-31, tentacles were mostly kept open and most of peristalses travelled upward.

The irregularities seen in monopolypids P-31, P-32 and P-33 may be attributable to their age. The monopolypids P-34 and P-35 were 19 to 20 days old, while P-31,

P-32 and P-33 were 22 to 23 days old and seemingly near the end of their surviving time under the unfed condition. Actually these three monoplypids showed a relatively clear peristaltic rhythm when they were 9 to 16 days old as seen in Table 2. Therefore, it may be concluded that a relation between the state of tentacles and the peristaltic direction is maintained only in younger vivid monoplypids under the unfed condition.

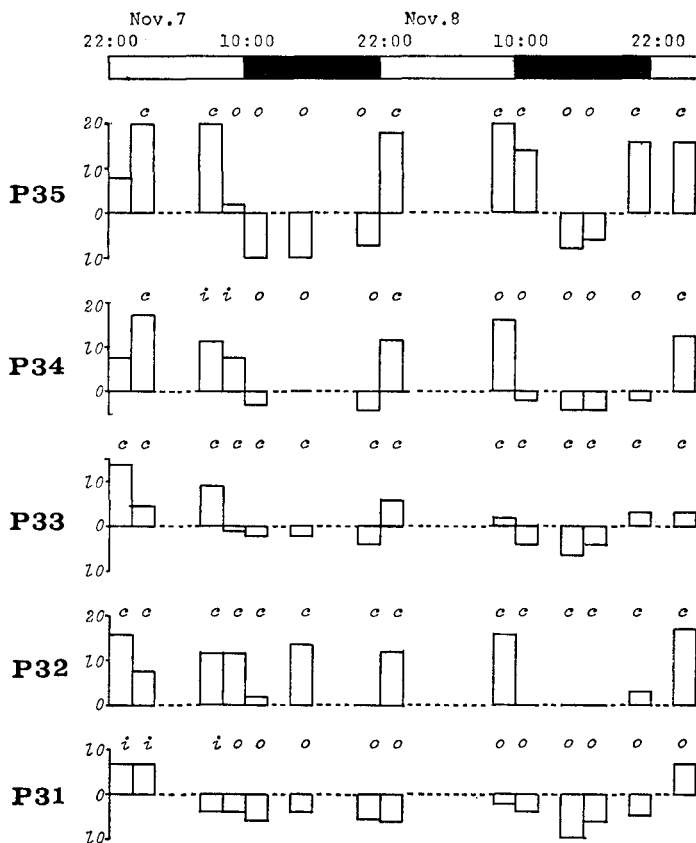


Fig. 8. Relation between the peristalsis and state of tentacles. The histogram shows the number of peristalses per 30 minutes; the downward peristalsis above and the upward peristalsis below the base line. States of tentacles are shown as *c* (closed) *o* (open), and *i* (intermediate state). The light and dark (dim light) phases are given at the top of the figure.

V. Effect of pH.

The diurnal alternation of direction in the peristalsis of the monoplypoid bodies is thought to be related with the expansion-contraction rhythm of adult colonies; contraction waves travelling from the peduncle to the top of the rachis in expansion but from the top of the rachis to the peduncle in contraction. As the upward peristaltic movement in adult colonies can be seen when the expansion is induced artificially in the daytime by decreasing pH of the body fluid, it was tried to see whether

the change of pH in the medium affects the frequency or direction of the peristalsis of monopolypids.

V-1. On the peristalsis induced by light.

The monopolypid maintaining the peristalsis induced by the light of 5000 lux was exposed to the sea water with pH modified by adding the sea-water solutions of carbon dioxide or sodium hydroxide. The addition of the acid or more basic sea water into the vessel water was made carefully lest the water turbulence disturbed the monopolypids. But, in returning to the former state the whole medium was replaced with the normal sea water, and so the animals were obliged to meet to a great and sudden disturbance.

A monopolypid body (P-28) of 17 days old was exposed to the sea water acidified to different degrees. The frequency of the downward peristalsis was maintained almost unchanged at pH 7.85 and 7.42, but at 7.18 it decreased a little with the upward peristalsis intervened once (Fig. 9); it is unknown whether the last mentioned was due to the pH change. When the pH was dropped to 7.00, the monopolypid continued several downward peristalses for a short time but then stopped all movements, closing its tentacles and contracting the body strongly; this caused the peduncle surface becoming rugged. Such features were observed in the sea water less acidified, too, though unremarkable. The replacement of the whole acidified medium with the normal sea water seemed to stimulate animal very much, as the monopolypid recovered the downward peristalsis with a frequency higher than before, and continuing for a while after the pH was dropped again to 7.00, though the frequency was somewhat decreased.

In another monopolypid (P-22) of 17 days old exposed to the basic sea water, the frequency of the downward peristalsis was slightly changed with the rise of pH.

These experiments seemed to show that the acidified sea water affected the downward peristalsis suppressively while the more alkaline sea water acceleratedly.

V-2. On the peristalsis maintained under an artificial light-dark cycle without any other inducement.

Three monopolypids (P-25, P-26 and P-27) of 18 days old exhibiting peristalses under an artificial light-dark cycle with the light phase (2000 lux) for 12 hours from 10:00 to 22:00 and the dark phase of the dim light (30 lux), were exposed to the sea water acidified or more alkalized and observed in the light phase. The pH of the medium was adjusted the same as in the previous experiment. At the beginning of the experiment monopolypids P-25 and P-27 had been showing the downward peristalsis, while P-26 the upward peristalsis even in the light phase.

When pH was dropped from 8.19 to 6.56, the downward peristalsis was extremely suppressed in P-25 and P-27, but P-26 maintained the upward peristalsis steadily but with a slightly decreased frequency. When the pH of the medium was restored to former 8.19, the three animals showed similarly the downward peristalsis of higher frequencies. This might be attributable to the mechanical stimulation of changing the whole vessel water. After one hour, P-25 and P-27 were found maintaining the downward peristalsis of relatively high frequencies, but P-26 was found having

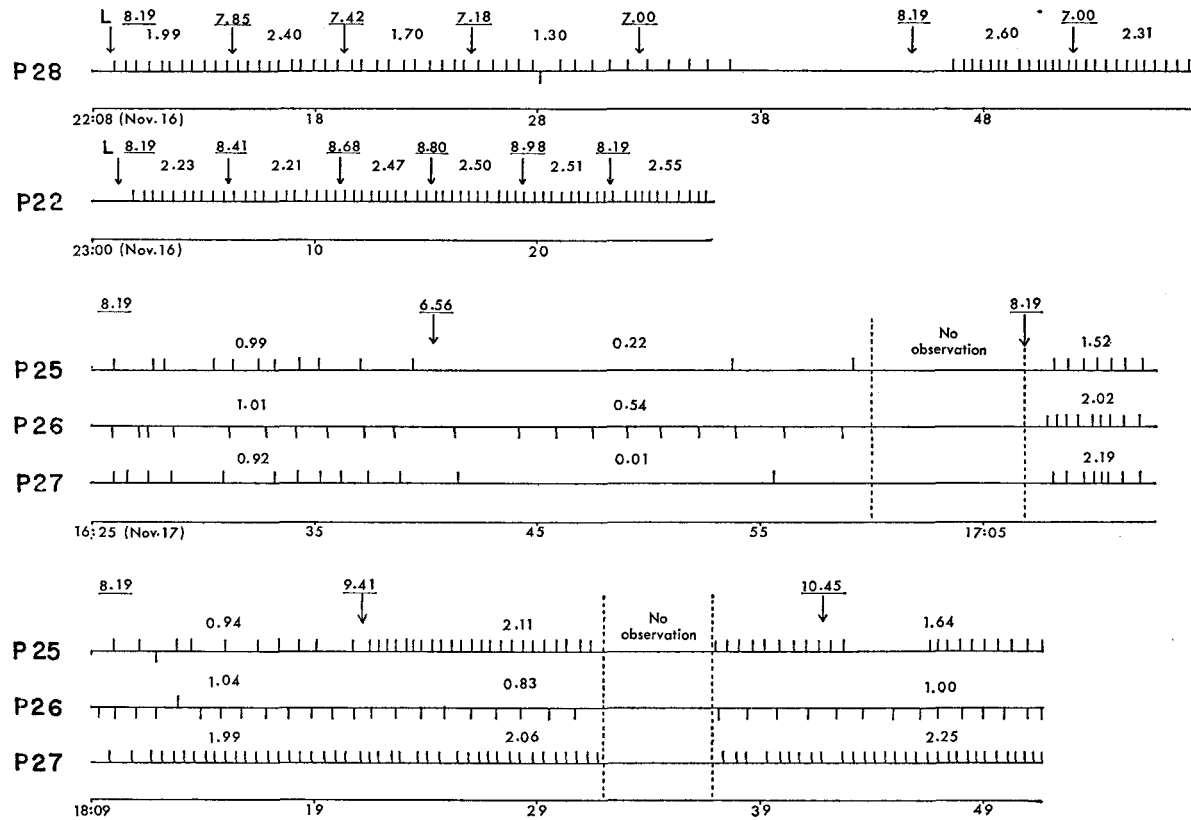


Fig. 9. Effect of pH. Monoplypid body P-28 subjected to the acidification of sea water, and monoplypid P-22 to the rise of alkalinity. Monoplypids P-25, P-26 and P-27 were subjected first to the acidification of sea water and then to the rise of alkalinity. The figures underlined indicate pH values and those at a little lower level and without underline show the frequency of peristalsis.

returned to the former upward peristalsis. Then the pH of the medium was raised to 9.41. Only the monopolypid P-25 reacted to this by raising the frequency of the downward peristalsis, but P-26 and P-27 kept their previous pace of former movements. At the pH rise to 10.45, monopolypid P-25 interrupted the movement for a short time, P-26 remained unchanged, and P-27 showed a slight increase of the frequency of its downward peristalsis.

These experiments show at least a tendency that the acidified sea water suppresses the downward peristalsis, while the sea water more alkalized accelerates the movement within a certain range. The upward peristalsis seemed nearly indifferent to pH change in P-26, but it is questionable whether this can be a general tendency, because maintaining of the upward peristalsis in the light phase itself is seemingly unnatural.

Discussion

I. *Peristalsis in the Monopolypid Stage.*

The peristalsis in juveniles of *Cavernularia* was found by Mori and Tanase (1973) in planulae older than 2 days old as well as monopolypid bodies. To learn more clearly in what condition the peristalsis occurs and by what kind of stimulation it is induced, close observations were made by the present author.

The first confirmed was that the downward peristalsis was induced by light. Then, the investigation of the relation between the light intensity and the frequency of the downward peristalsis (Fig. 3, Table 1) showed that the higher the light intensity was, the higher the frequency, as seen 2.4–2.6/min at 47700 lux, 1.7–1.8/min at 8000 lux (Fig. 3) and 1.0–1.5/min at 80–3400 lux (Table 1). Further, it was found that the change of the light intensity itself can be stimulative to induce the downward peristalsis and that in changing the intensity, the peristaltic frequency was always higher at a certain intensity when the light was raised to that intensity than at the same intensity but when the light was lowered to that level, as seen at 8000 (Fig. 3), 500 and 213 lux (Table 1). At lower intensities, however, the effect of light seemingly became obscure by unknown factors other than light.

The frequency of short rhythms are generally known to be affected by temperature change. And the peristalsis in monopolypid bodies of *Cavernularia* could not be exceptional. When the temperature was gradually raised, the temperature effect was most distinct in ranges of higher temperatures, for instance from 21.6 to 23.7°C (Fig. 5). On the other hand, the temperature rise or drop in the step method did not show so clear effects (Fig. 6). Comparing what were manifested by the curves shown in Fig. 6, it was suggested that the peristaltic frequency was temperature dependent.

All the results referred to above concern the downward peristalsis. There is known another type of peristalsis, the upward peristalsis. To see when the downward and upward peristalses were exhibited observations of monopolypids were made at different time of day (Figs. 7 and 8, Table 2). The downward peristalsis of higher frequencies was observed in the light phase, while the upward peristalsis was limited

to the dark phase. This is seemingly related to the peristalsis in adult colonies (see later), in which the expansion accompanying the rachidal peristalsis, the peristalsis travelling from the peduncle to rachis, was inducible by injection of the acidified sea water into the colony. If the upward peristalsis in the monopolydipids is really corresponding to the rachidal peristalsis in adult colonies, then acidified sea water might induce the upward peristalsis in the monopolydipids. Thus, the monopolydipid bodies were immersed in the acidified sea water, but they did not show any upward peristalsis, though the frequency of the downward peristalsis was affected by this treatment. This is probably attributable to the difference in treatment; pH of the body fluid was changed in adult colonies, while pH of the medium was varied in juveniles. The microinjection may be a better approach to this problem in future.

II. *Comparison of Peristalsis between Monopolydipid Bodies and Adult Colonies.*

Two types of the peristalsis are observed in the monopolydipid bodies of this sea-pen, the downward and upward peristalses, and also two kinds of peristalses have been reported in the natural state in the adult colonies, one travelling upward and the other downward (Mori, 1943). The upward peristalsis in adult corresponds to the rachidal peristalsis of Parker (1920), in *Renilla*, in which contraction waves travel from the distal end of the peduncle to the rachidal part. The downward peristalsis in adult is called "the peduncular peristalsis" in the present paper, extending the Parker's definition a little, in which this is limited to the peristalsis travelling from the proximal to the distal part of the peduncle.

As the rachidal peristalsis is seen in the expansion and the peduncular peristalsis in the contraction of adult colonies and the sea-pen expands from dusk to midnight and contracts from midnight to dawn, the two types of peristalses are alternating regularly under the natural conditions. In the monopolydipid bodies, either expansion or contraction is not yet seen but only the diurnal alternation of direction of the peristalsis. Therefore, the upward and downward peristalses in juveniles is regarded to represent the primitive state of the rachidal and peduncular peristalses in adult colonies.

In the rachidal peristalsis in adult colonies, contraction waves travel from the distal end of the peduncle to the middle or distal part of the rachis, while in the upward peristalsis in monopolydipid bodies the waves travel from the distal end of the peduncle to the region just above the buds of secondary polyps or to the base of tentacles (Fig. 2). In the peduncular peristalsis in adult colonies, contraction waves are started at the distal end of the rachis and travel down to the proximal part of the rachis or to the distal end of the peduncle, though the deceasing point of waves has not been observed on the colonies standing naturally, because the peduncle is burried in the sand in that state. On the colonies laid in the glass vessel, wholly exposed, the waves are observed to travel to the distal part of the peduncle (Honjo, 1940). In the downward peristalsis in monopolydipid bodies, the waves travel from the precursory rachidal part near the secondary buds to the distal end of the peduncle (Fig. 1). Thus, the contraction waves travel, in both the upward and downward peristalses in monopolydipid bodies,

the ranges corresponding respectively to those travelled by the waves in the rachidal and peduncular peristalses in adult colonies.

The travelling rate of contraction waves is different between the monopolypid juvenile and adult. It is 0.2 to 1.2 cm/min for the upward and 0.2 to 1.6 cm/min for the downward peristalsis in monopolypid juveniles, while it is 48 to 72 cm/min for the rachidal and 30 to 48 cm/min for the peduncular peristalsis in adult colonies (Honjo, 1940). In adult colonies of *Renilla*, it is 72 cm/min and 66 cm/min for the rachidal and peduncular peristalsis, respectively (Parker, 1920).

The frequency of the peristalsis is affected by light intensity, temperature and pH of the medium and is different, as seen in the present experiments, according to the cycle phase and further to the exact time of observation. It is 0 to 24 waves/30 min for the upward and 0 to 44 waves/30 min for the downward peristalsis in the monopolypid bodies kept under the light-dark cycle. It is 10 to 20 waves/30 min and 50 waves/30 min respectively for the rachidal and peduncular peristalses in adult colonies of *Renilla* (Parker, 1920).

As mentioned above, in adult colonies, the peristalsis is closely correlated with the expansion-contraction behaviour, the rachidal peristalsis always observable at the expansion of the colony and the peduncular peristalsis at the contraction. This is true when the expansion is induced by injection of the acidified sea water. Further, the expansion of adult colonies occurs always in the dark time and thus is related with darkness, while the contraction with the lighted condition, as the contraction of an expanded colony in darkness can be induced by illumination. On the other hand, in the monopolypid bodies, the upward peristalsis is nearly limited to the dark phase, while the downward peristalsis is performed prominently in the light phase. The upward peristalsis maintained in the dark phase may be reversed to the downward peristalsis by illumination (unpublished data).

From these observations, it is concluded that the upward peristalsis in monopolypid bodies represents the primitive state of the rachidal peristalsis in adult colonies and the downward peristalsis in monopolypids corresponds to the peduncular peristalsis of grown-up colonies. The monopolypid juveniles of this sea-pen do not show any daily expansion-contraction behaviour, but the diurnal alternation of direction in the peristalsis.

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REFERENCES

- Brafield, A.E. 1969. Water movements in the pennatulid coelenterate *Pteroides griseum*. J. Zool. London, 158, 317-325.
- Buisson, B. 1971. Les activités rythmiques comportementales de la colonie de *Veretillum cynomorium* (Cnidaire Pennatulidae). Cah. Biol. Mar., 12, 11-48.
- Ceccatty, M.P. and Buisson, B. 1965. Reciprocal behavior of the rachis and peduncle in colonies of *Veretillum cynomorium* Pall. Amer. Zoologist, 5, 531-535.
- Honjo, I. 1940. Beiträge zur Nervenmuskelphysiologie der koloniebildenden Tiere. I. Die Peristaltik von *Cavernularia*. Annot. zool. Jap., 19, 301-308.
- Imafuku, M. 1973. On some physiological aspects in the daily rhythmic activity of the sea-pen, *Cavernularia obesa* Valenciennes. Publ. Seto Mar. Biol. Lab., 20 (Proc. Second Internat. Symp. Cnidaria), 431-454.
- Miyajima, M. 1897. Ecological observations of a sea-pen (*Veretillum*). Zool. Mag., 9, 367-371. (In Japanese).
- . 1900. A sea-pen (*Cavernularia obesa* Val.). Ibid., 12, 426-433. (In Japanese).
- Mori, S. 1943. Daily rhythmic activity of the sea-pen, *Cavernularia obesa* Valenciennes. I. Observations in nature. Ibid., 55, 285-291. (In Japanese).
- Mori, S. and Tanase, H. 1973. Studies on the daily rhythmic activity of the sea-pen, *Cavernularia obesa* Valenciennes. XVIII. Ontogenetic development of the daily rhythmic activity. Publ. Seto Mar. Biol. Lab., 20 (Proc. Second Internat. Symp. Cnidaria), 455-467.
- Parker, G.H. 1920. Activities of colonial animals. II. Nenromuscular movements and phosphorescence in *Renilla*. J. Exp. Zool., 31, 475-515.
- Wilson, E.B. 1883. The development of *Renilla*. Philos. Trans., Roy. Soc., London, 174, 723-815.