PHYSIOLOGICAL STUDIES ON
MARPHYSA GRAVELYI SOUTHERN

II. Volume Regulation*

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1. INTRODUCTION

In an earlier paper (Krishnamoorthi and Krishnaswamy, 1966a) it was shown that Marphysa gravelyi Southern, an Eunicid polychaete of the Adayar brackish water zones, tolerated lowered salinities over a wider range—5% to 28%—than any other polychaete studied from the same region, viz., Glycera embranchiata, Onuphis eremita, Loimia medusa, Clymene insecta (= Euclymene insecta Hartman) (Krishnamoorthi, 1962) and Diopatra variabilis (Krishnamoorthi, 1963b). Discussing the obvious advantage gained by Marphysa gravelyi in this regard over the other polychaetes of that region, it was argued that endurance of reduced salinities alone, unaccompanied by matching abilities of volume control, would be of little advantage, since polychaetes invading zones of fluctuating salinities are known to swell followed by varying degrees of volume regulation (Schlieper, 1929a and b; Beadle, 1931, 1937; Sayles, 1935; Ellis, 1937, 1939; Topping and Fuller, 1942; Smith, 1955c, 1959; Jorgensen and Dales, 1957). The present paper is an attempt to evaluate experimental results on the capacities of M. gravelyi over regulation of volume under stresses of hyposmotic media.

2. MATERIAL AND METHODS

The method of collection of Marphysa gravelyi Southern from the brackish water zone of Adayar, Madras, and the procedure of experimentation, were similar to those followed earlier (Krishnamoorthi, 1962, 1963b; Krishnamoorthi and Krishnaswamy, 1966a). All experiments were conducted

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at the room temperature of 29.5 ± 0.5 °C. Volume changes were followed by the displacement method of weighing aquatic animals using specific gravity bottles (Lowndes, 1942). A specific gravity bottle of capacity 25 ml. was found quite suitable. Since preliminary experiments had shown that there was no appreciable change in volume beyond a period of 24 hours, observations further to this period were discontinued and the experiments terminated. All values reported here are the means of 10 determinations.

3. Results

3.1. Volume regulation over a period of 24 hours in a hyposmotic medium of strength 6%

The volume in all the worms reached the maximum percentage increase (40.49%) at the end of 1st hour (Table I, Fig. 1). Thereafter there was a steady fall reaching the lowest volume, viz., 4.29% at the end of the 4th hour. Beyond the 4th hour, intermittent increase and decrease in the volume were noticed. But the increase was never as high as that observed during either the 1st or the 2nd or the 3rd hours when the percentage increases were 40.49, 31.90, 12.27 respectively. The initial increase in volume is, perhaps, due to absorption of water from the medium against an osmotic gradient and the subsequent fall a consequence of loss of sats as reported in a number of

![Graph showing volume regulation over a period of 24 hours in a hyposmotic medium of strength 6%.](image-url)
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<table>
<thead>
<tr>
<th>Volume in c.c. (mean of 10 determinations) after hourly exposure to experimental medium of salinity</th>
<th>Mean</th>
<th>s.d.</th>
<th>s.e.</th>
<th>% increase</th>
<th>Mean</th>
<th>s.d.</th>
<th>s.e.</th>
<th>% increase</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial</td>
<td>1.63</td>
<td>0.36±0.120</td>
<td>0.020±0.016</td>
<td>1.8</td>
<td>0.78±0.025</td>
<td>36.13</td>
<td>15.81</td>
<td>5.36</td>
</tr>
<tr>
<td>1</td>
<td>2.29</td>
<td>0.26±0.090</td>
<td>0.032±0.010</td>
<td>2.2</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
<tr>
<td>2</td>
<td>2.15</td>
<td>0.022±0.007</td>
<td>0.020±0.007</td>
<td>2.1</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
<tr>
<td>3</td>
<td>1.83</td>
<td>0.080±0.030</td>
<td>0.020±0.007</td>
<td>1.8</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
<tr>
<td>4</td>
<td>1.70</td>
<td>0.220±0.017</td>
<td>0.020±0.007</td>
<td>1.7</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
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<td>5</td>
<td>1.67</td>
<td>0.250±0.017</td>
<td>0.020±0.007</td>
<td>1.6</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
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<td>6</td>
<td>1.71</td>
<td>0.270±0.017</td>
<td>0.020±0.007</td>
<td>1.7</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
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<td>7</td>
<td>1.77</td>
<td>0.400±0.130</td>
<td>0.020±0.007</td>
<td>1.8</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
<tr>
<td>8</td>
<td>1.71</td>
<td>0.090±0.030</td>
<td>0.020±0.007</td>
<td>1.7</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
<tr>
<td>23</td>
<td>1.71</td>
<td>0.090±0.030</td>
<td>0.020±0.007</td>
<td>1.7</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
<tr>
<td>24</td>
<td>1.68</td>
<td>0.080±0.030</td>
<td>0.020±0.007</td>
<td>1.7</td>
<td>0.71±0.020</td>
<td>36.24</td>
<td>15.86</td>
<td>5.40</td>
</tr>
</tbody>
</table>

**Table I**

Volume changes after every hour of exposure over a period of 24 hours to hypoosmotic media of salinity 6%, 8%, and 13%. Temperature: 29.5 ± 0.5°C.
polychaetes, viz., *N. diversicolor*, *N. pelagica*, *N. virens*, *Perinereis cultrifera* (Schlieper, 1929a and b; Beadle, 1931, 1937; Sayles, 1935; Jørgensen and Dales, 1957); in *N. limnicola* (= *Neanthes lighti*) (Smith, 1956, 1959); in *Onuphis eremita*, *Loimia medusa* and *Clymene insecta* (= *Euclymene insecta* Hartman) (Krishnamoorthi, 1962) and in *Diopatra variabilis* (Krishnamoorthi, 1963b). The oscillations in the volume after the 4th hour, are, perhaps, individual efforts of the worm to keep down the increase in volume. Also it is seen that the final volume attained at the end of the 4th hour and maintained till the 24th hour, is always higher than the original volume. The final volume ranged between 2.45% to 8.59%.

3.2. Volume regulation over a period of 24 hours in a hyposmotic medium of strength 8%.

It may be seen that in this salinity also as in the previous hyposmotic medium, all the worms swelled to the maximum of 36.13% at the end of the 1st hour and subsequently decreased in volume till the end of the 4th hour, the percentage increase in volume noted at the end of the 4th hour being 2.58% (Table I, Fig. 2). It steadily increased to 9.03% till the end of the 7th hour, only to fall to 2.58% at the end of the 24th hour. In other words, the final volume reached at the end of the 4th hour up to the 24th hour, must be due to similar factors as that prescribed for the behaviour of the worms.
in the previous experiment. However, one observation is irresistible, i.e., the final volume attained at the end of the 4th hour and maintained at that volume till the end of the 24th hour is not only higher than the original volume, similar to the behaviour of the worms in the previous salinity, but more or less of the same magnitude as that attained in the previous hyposmotic medium.

3.3. Volume regulation over a period of 24 hours in a hyposmotic medium of strength 13%

In this medium also the responses of the worms were not dissimilar from those observed in the two previous media (Table I, Fig. 3). They attained the maximum volume of 25.60% at the end of the 1st hour; and as in the previous two salinities, the volume subsequently decreased reaching the minimum of 1.19% at the end of the 4th hour. Thereafter, till the end of the 24th hour, the volume fluctuated between 1.19% and 6.55%, the latter percentage increase registered at the end of the 6th hour. The same factors are, perhaps, applicable to explain the responses of the worms observed in the present experiment too. However, while the final volume fluctuated in the present medium between 1.19% and 5.36%, the corresponding ranges were higher in the two previous media.

![Figure 3](image_url)

**Fig. 3.** Indicating changes in volume after every hour of exposure to a hyposmotic medium of 13% over a period of 24 hrs. Temp.: 29.5 ± 0.5°C. Each point is the mean of 10 determinations.
4. Remarks

In all the experiments the behaviour of *Marphysa gravelyi* was consistently similar. It attained the maximum volume at the end of the 1st hour and subsequently decreased to the lowest value observed at the end of the 4th hour. Thereafter, it was characterised by intermittent rises and falls till the end of the experiments, i.e., 24 hours. *M. gravelyi*, thus, resembles *Nereis diversicolor* and *Nereis pelagica* (Schlieper, 1929 a and b); *N. diversicolor* and *Perinereis cultrifera* (Beadle, 1931, 1937); *Nereis virens* (Sayles, 1935); *N. virens*, *N. pelagica* and *N. diversicolor* (Jørgensen and Dales, 1957) and *Nereis limnicola* (Smith, 1959) in its response to hyposaline media. However, even in this similarity of pattern, there is yet a difference apparent. The magnitude of increase in volume at the end of the 1st hour as well as the final volume attained and maintained from the 4th to 24th hour were different in different hyposaline media. While the maximum increase in volume in hyposaline media of strength 6%o was 40·49%, the respective increases in volume in media of salinities 8%o and 13%o were 36·13% and 25·60%. Similarly, the final volumes reached in sea-water diluted to have a concentration of 6%, 8%o and 13%o were 4·29%, 2·58% and 1·19% respectively. In other words, both the initial increase in volume and the final volume attained are, perhaps, a function of the dilution of the experimental media. Once again, the responses of *M. gravelyi* to the stresses of hyposaline media, are, thus, not far different from those observed in *N. diversicolor*, *N. virens*, *N. pelagica*, *Perinereis cultrifera* and *N. lighti* (Schlieper, 1929 a and b; Beadle, 1931, 1937; Sayles, 1935; Jørgensen and Dales, 1957; Smith, 1959). But the magnitude of change in volume under identical stresses of an hypo-osmotic medium, was more in *N. virens* than in *N. diversicolor* (Schlieper, 1929 a and b). *P. cultrifera* swelled considerably compared with *N. diversicolor* (Beadle, 1931), while *N. virens* ranked between *N. pelagica* and *N. diversicolor* (Jørgensen and Dales, 1957). The extent of volume change in *M. gravelyi* therefore, would be significant only when compared (Table II) with similar parameters obtained from the responses of other polychaetes, viz., *G. embranchniata*, *O. eremita*, *L. medusa* and *C. insecta* (Krishnamoorthi, 1962) and *D. variabilis* (Krishnamoorthi, 1963 b) that co-exist with *M. gravelyi* in the Adyar estuary. It has already been shown that volume control in *C. insecta* was better developed than in either *G. embranchniata* or *L. medusa* or *O. eremita*, since the magnitudes of increase in volume and the final volume attained at the end of the experiment were the lowest compared with those of *L. medusa* and *O. eremita* (Krishnamoorthi, 1962). Between *C. insecta* and *D. variabilis*, the latter exhibited better capacities for volume regulation (Krishnamoorthi,
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TABLE II

Statement of comparative performance of volume control among 6 species of polychaetes inhabiting the brackish-water zones of Adyar, Madras

<table>
<thead>
<tr>
<th>Name of species</th>
<th>% increase of volume after 1 hour of exposure to dilutions of salinities (%)</th>
<th>% final volume after 4 hours of exposure to dilutions of salinities (%)</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>6 8 13 20</td>
<td>6 8 13 20</td>
<td></td>
</tr>
<tr>
<td>O. eremita</td>
<td>60.0 44.7 29.4</td>
<td>18.1 17.4 12.6</td>
<td>Krishnamoorthi, 1962</td>
</tr>
<tr>
<td>L. medusa</td>
<td>58.6 42.2 26.8</td>
<td>17.6 15.6 10.3</td>
<td></td>
</tr>
<tr>
<td>C. insecta</td>
<td>50.8 35.8 21.1</td>
<td>10.4 8.2 5.2</td>
<td></td>
</tr>
<tr>
<td>G. embranchiata</td>
<td>55.0 40.0 28.0</td>
<td>60.0 45.5 32.3</td>
<td></td>
</tr>
<tr>
<td>D. variabilis</td>
<td>50.0 40.0 28.0</td>
<td>10.0 8.0 5.0</td>
<td>Krishnamoorthi, 1963 b</td>
</tr>
<tr>
<td>M. gravyyi</td>
<td>40.5 36.1 25.6</td>
<td>4.3 2.6 1.2</td>
<td>Present Study</td>
</tr>
</tbody>
</table>

1963 b). Comparing the increase in volume of D. variabilis with that of M. gravyyi in hyposmotic media of salinities 8% and 13%, it would be seen that, whereas the percentage increase in volume at the end of the 1st hour in the respective dilutions were 50 and 40 for D. variabilis, similar figures for M. gravyyi were only 36 and 26. Also the final volumes attained were lower in M. gravyyi than in D. variabilis. In M. gravyyi, therefore, the mechanism of volume regulation is far superior to that obtained in other forms. Observing lesser degree of swelling in N. limnicola obtained from Lake Merced than in those from the estuary, Walker Creek, Smith (1959) suggested better volume control in the former population. Volume regulation, therefore, has constituted an effective mechanism enabling M. gravyyi withstand changes, however moderate or big, in the salinity of the medium they inhabit.

The increase in volume in M. gravyyi is, probably, due to absorption of water against an osmotic gradient, in agreement with the observations of earlier workers in the field of physiology of Nereids. Could the observed differences in the magnitude of increase in volume, be a reflection of the extent of permeability? Jørgensen and Dales (1957) following the volume
changes in three species of Nereids, *viz.*, *N. virens*, *N. pelagica* and *N. diversicolor*, suggested and later proved with experiments involving use of radioactive isotope, $^{36}$Cl, that the lower values of increase in volume in *N. diversicolor* could only be due to it being less permeable than either *N. Virens* or *N. pelagica*. Among the polychaetes studied (Krishnamoorthi, 1962, 1963 b), *M. gravelyi* shows the lowest rate of increase in volume and the final volume attained is also lower. In the light of recent evidences let in (Smith, 1963 b) that quicker rate of loss of salts may also be advantageous, a reduction in permeability coupled with the above factor may considerably increase the chances of quicker establishment of a species in a changing environment. Although similar studies in *M. gravelyi* must await future investigation, the observed rate of increase in volume being comparatively lower, it is conceivable that in *M. gravelyi* also the rate of permeability is, perhaps, the lowest.

Factors like tolerance of salinity over a wider range; greater capacities for volume regulation and reduction in the permeability, admirable and meaningful as they are, for a successful penetration into and establishment of a polychaete such as *M. gravelyi* in brackish waters, fail to answer the question of maintenance of a constant internal concentration, a factor equally profitable and necessary to meet the hazards of a changing environment. If it is assumed that the initial increase in volume in *M. gravelyi* is due to absorption of water against an osmotic gradient and the subsequent decrease a consequence of loss of salts, such an assumption presupposes profound changes in the body fluids, any imbalance of which would be disastrous. It appeared, therefore, that the next stage in the course of the present investigations, should be directed to the study of body fluids and its consequential changes as reflected by the depression in the freezing point, under stresses of heterosmotic media.

5. SUMMARY

Like all other polychaetes, *M. gravelyi* also swells when exposed to hypotonic media. But the extent of increase was considerably low. In this worm, as in other polychaetes, the initial increase and the final volume attained are a function of the external medium. Both the final volume and the initial increase being very low in *M. gravelyi*, reduction in permeability is, perhaps, the maximum.

6. REFERENCES

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Ellis, W. G. “The water and electrolyte exchange of Nereis diversicolor (Muller),” Ibid., 1937, 14, 340-50.


Topping, F. L. and Fuller, J. F. “The accommodation of some marine invertebrates to reduced osmotic pressures,” Ibid., 1942, 82, 372-84.