On the Voluminal Relation Between the Ectoplasm and Endoplasm in a Heliozoon, *Actinosphaerium eichhorni* (Ehrenberg)

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I. Introduction

According to HUXLEY (1924, 1932), SCHMALHAUSEN (1927) and others, the relative growth in Metazoa can generally be expressed by the equation $y = bx^k$, where x and y are the measurements, b the fractional coefficient, and k the specific exponent. It is the purpose of the present investigation to find whether this relation holds in the Protozoa as well, in other words, whether the growth of a unicellular organism accords with the same rule as that for a multicellular metazoan body.

Thus far the growth of the Protozoa has been studied either in linear dimension (PEARL 1907, JENNINGS 1908, SCHMALHAUSEN 1925) or in areal dimensions (POPOFF 1909, MIZUNO 1927) only, and no work has been done, except that of CHALKLEY (1931), on the growth in cubic dimensions, although apparently by such a work alone the actual phase of growth of a cell can be obtained.

The heliozoon, *Actinosphaerium eichhorni* is a material fitted for such a study, because of its relatively large size and very regular spherical form.

Before going further, I wish to express my sincere thanks to Prof. TAKU KOMAI for his generous guidance and also for his kindness in revising the manuscript.

II. Material and Methods

The stock was derived from the material collected by Mr. B. OSOGOE from Lake Biwa, and has been cultivated in the laboratory since April, 1937. The standard culture medium is as follows: 50 cc. of water from a pond near the laboratory are poured in a PETRI dish 10 cm. in diameter and two grains of rice are added to it, and the dish is kept at 18°–21°C. in a thermostat.

Several pure lines have been isolated, but so far no genetical diversity in growth has been observed among them.

The body of this heliozoon is practically a regular sphere and the optical cross-section shows two concentric circles. The inner circle limits the endoplasm and the outer one the ectoplasm. If Dc represents the diameter of the outer circle, and Dn that of the inner one, then the volumes of the ectoplasm and endoplasm can be calculated as follows:—

$$\text{Volume of endoplasm } (x) = Dn^3 \frac{\pi}{6} u^3 \mu^3,$$

$$\text{Volume of ectoplasm } (y) = (Dc^3 - Dn^3) \frac{\pi}{6} u^3 \mu^3,$$

where u is the unit of the ocular micrometer ($u = 10.5 \mu$), $\frac{\pi}{6} u^3 \mu^3$ ($= 193\pi \mu^3$) being the unit of the calculated volume. Both Dn^3 and $Dc^3 - Dn^3$ are the actual numbers which are important in analysing the voluminal relation. In this computation the volume of the axopodia which consist of ectoplasm are disregarded, as this seems to be negligible for the estimation of the entire volume of the ectoplasm.

III. Analyses of the Voluminal Relation

1. Voluminal relation in the normal individuals

a). *Culture No. 12.*

This culture was started on May 25, 1937, and the measurements were made on June 8, 1937. The diameters of the ectoplasm and endoplasm of 250 individuals were measured, and from the values thus obtained the protoplasmic volumes of the individuals were computed (Table 1)*. If the logarithms of the volumes of the ectoplasm are plotted against those of the volumes of the endoplasm, a straight line may be obtained (fig. 1). The linear equation is

$$\log y = \log b + k \log x,$$

where b and k are constants, and x and y are the values of the endoplasm and ectoplasm respectively. From this straight line, the values of k and b can be determined graphically,

$$k = 0.72,$$

$$b = 21.78.$$

Then the straight line is $\log y = \log 21.78 + 0.72 \log x$,

$$\text{so} \quad y = 21.78 x^{0.72}. \quad (\text{I})$$

* For the economy of space, the other tables are not published.

Table 1. Culture No. 12

Diameter of ectoplasm in 10.5μ D_c	Diameter of endoplasm in 10.5μ D_n	Number of individuals	Volume of endoplasm in $193\pi\mu^3$ $Dn^3 = x$	Volume of ectoplasm in $193\pi\mu^3$ $Dc^3 - Dn^3 = y$
20	12.5	4	1950	6050
21	14.3	3	2920	6330
22	14.7	11	3180	7350
23	15.9	8	4020	8160
24	16.5	18	4490	9330
25	17.4	20	5270	10360
26	18.4	22	6230	11350
27	19.0	35	6860	12820
28	19.8	38	7660	14290
29	20.5	16	8620	15770
30	21.6	23	10010	16990
31	22.5	17	11390	18400
32	23.1	17	12430	20340
33	24.3	7	14350	21590
34	25.4	5	16390	22920
35	25.3	3	16190	26680
36	27.5	1	20800	25600
41	31.0	1	(29790)	(39130)
		250		

The voluminal relation between the ectoplasm and endoplasm can be expressed by this equation I (fig. 2).

Fig. 1. Logarithms of the volumes of the ectoplasm plotted against the logarithms of the volumes of the endoplasm.

Fig. 2. Voluminal relation between the ectoplasm and endoplasm in normal individuals. The unit of the volume is $193\pi\mu^3$.

b). Culture No. 13.

This culture was started on May 25, 1937, and the measurements were made on June 15, 1937. The values of the measurements of

Fig. 3. Voluminal relation between the ectoplasm and endoplasm in normal individuals. The unit of the volume is $193 \pi \mu^3$.

300 individuals were analyzed in the same manner, and the equation II was obtained (fig. 3):

$$y = 32.36 x^{0.68} \quad (\text{II}).$$

c). *Culture No. 80.*

This culture was started on October 21, 1937, and the measurements were made on November 18, 1937. From the values of the measurements of 200 individuals, the equation III was obtained (fig. 3):

$$y = 26.00 x^{0.71} \quad (\text{III}).$$

2. Influence of the artificial plasmotomy on the voluminal relation

The cell-body of *Actinosphaerium* is easily divisible into two parts by a glass capillary. Each piece becomes a small but complete animalcule in two or three hours. This fact has been confirmed by several investigators (OKADA 1930). The influence of such plasmotomy upon the voluminal relation between the ectoplasm and endoplasm was studied.

a). *Culture No. 80.*

This culture was started on October 21, 1937, and the operations were made on November 15, 1937. 15 mother individuals were operated on and resulted in 30 daughter individuals. The equation IV was determined from these individuals:

$$y = 21.73 x^{0.73} \quad (\text{IV}).$$

The values for the mother and daughter individuals plotted, show clearly that the voluminal relation of all the 45 individuals may be expressed by the same equation IV (fig. 4).

b). *Culture No. 88a.*

This culture was started on December 22, 1937, and the operations were made on March 23, 1938. 20 mother individuals were operated on and 40 individuals obtained:

$$y = 38.91 x^{0.65} \quad (V).$$

As shown in fig. 4, the values for both the mother and daughter individuals may be expressed by the equation V

Fig. 4. Voluminal relation between the ectoplasm and endoplasm in mother and daughter individuals produced by artificial plasmotomy. The equation determined for the daughter individuals satisfies that for the mother individuals.

The unit of the volume is $193 \pi \mu^3$.

Fig. 5. The change of the voluminal relation according to medium. In ordinary pond water b becomes larger, and k smaller than in old culture medium. The unit of the volume is $193 \pi \mu^3$.

3. Influence of ordinary pond water on the voluminal relation

As shown in the foregoing paragraphs, the voluminal relation can be given by the equation $y = bx^k$. In this equation, are both b and k constant for each individual, or do they vary according to environmental conditions? The following experiment was performed to answer this question.

a). *Culture No. 88a.*

40 daughter individuals obtained in the foregoing experiment were kept in ordinary pond water for 24 hours at 18°C . (March

23–24, 1938). In the mean time, some of these individuals had divided and the number increased to 54. The measurements of these individuals have given the equation VI (fig. 5) :

$$y = 113.50 x^{0.57} \quad (\text{VI}).$$

b). *Culture No. 105.*

This culture was started on December 22, 1937, and the experiment was made on March 21–22, 1938. 40 individuals were kept in ordinary pond water for 24 hours at 18°C. The equation VII for these individuals was determined prior to the change of medium :

$$y = 28.19 x^{0.71} \quad (\text{VII}).$$

By the following day, the number of individuals had become 55. The measurements of these individuals have given the equation VIII (fig. 6) :

$$y = 51.29 x^{0.65} \quad (\text{VIII}).$$

Fig. 6. The change of the voluminal relation in ordinary pond water. The same result as shown in fig. 5 has been obtained.

individuals kept in ordinary pond water than in those in the culture medium.

IV. General Consideration

It has been shown above that the voluminal relation between the ectoplasm and endoplasm of *Actinosphaerium eichhorni* can be expressed by the equation $y = bx^k$, as in the case of the Metazoa. In this equation x and y are the measurements, b the fractional coefficient, and k the specific exponent (HUXLEY and others). HUXLEY (1932) is of opinion that b is "of no particular biological significance",

while NOMURA (1927) has pointed out for the growth of molluscan shells that b may be a local constant.

As may be seen in equations I–VIII above, both b and k are of no fixed value for the whole population of *Actinosphaerium*, each culture having a definite set of b and k . The values of b and k thus seem to be determined by the conditions in the culture medium in which the organisms are kept. Pure lines seem to have little significance for the variation of these constants.

In the daughter individuals produced by artificial plasmotomy, the protoplasmic differentiation and the restitution of lost parts are accomplished in strict accordance to the original state in the mother individuals. In other words, a definite voluminal relation between the ectoplasm and endoplasm seems to be of absolute necessity for the maintenance of the life of this heliozoon.

Further, it has been shown that the change of the medium from the culture medium to ordinary pond water results in the change of the voluminal relation. Thus the voluminal relation is easily changeable in accordance with environmental conditions. In ordinary pond water, b tends to become larger and k smaller, than in the old culture medium.

V. Summary

1) The voluminal relation between the ectoplasm and endoplasm in the heliozoon, *Actinosphaerium eichhorni* (EHRENBERG) can be given by the same equation $y = bx^k$ as in the case of the organs of the Metazoa, where b and k are constants, x the volume of the endoplasm, and y the volume of the ectoplasm.

2) The voluminal relation in daughter individuals produced by artificial plasmotomy is the same as that in the mother individuals prior to the operation.

3) All the individuals in the same culture dish take one definite set of b and k at a given moment.

4) The values of b and k , however, are not fixed, but fluctuate within a wide range according to the environmental condition. In ordinary pond water, the value of b is larger, and that of k smaller than in the old culture medium.

VI. Literature

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