in different substrate concentrations was compared by a method similar to those described for examination of pH effect. These results are presented in Figure 2. The average value of Michaelis constant is $1 \cdot 10^{-3}$ [7]. To examine the effect of Mg^{++} ions, the OPD increase of the same cells was compared during incubation in media with and without magnesium ions. Contrary to the properties of alkaline phosphatase in kidney brush border, 15% activatory effect was noted[3].

As a preliminary experiment for further studies on the effect of different fixatives on the enzymatic activity, the influence of formaline was examined. The incubation and measurement of OPD in optimal incubation medium were performed after ordinary fixation in cold acetone, and after removal of the calcium phosphate the chamber was filled with 10% cold neutralized formaline and kept in a refrigerator at 4°C for 2 h. Then the formaline was rinsed out, the same incubation medium was introduced, and the enzymatic activity of the same cells was measured. It was found that the activity was lowered by about 55% after formaline treatment.

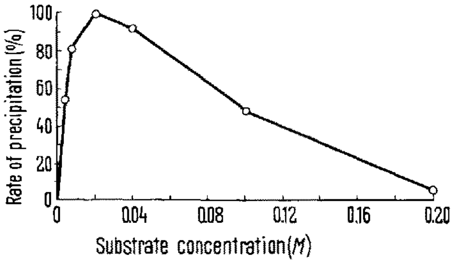

Fig. 2

Zusammenfassung. Mittels Interferenzmikroskopie wurde die Präzipitationsrate des Calciumphosphats in Ehrlich Ascites-Tumorzellen gemessen und die Beeinflussung der alkalischen Phosphataseaktivität durch diverse Faktoren untersucht.

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Department of Histology and Embryology, School of Medicine, Warsaw (Poland), October 26, 1964.

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[7] H. LINEWEAVER and D. BURK, J. Am. chem. Soc. *56*, 658 (1934).

A Possible Way of Origin of Parthenogenetic Strains of *Dinophilus apatris* (= *D. gyrociliatus*)

Some fertilized females of *Dinophilus apatris* reproduce normally during most of their life, but at the end of their vital cycle they lay eggs which do not develop normally. The cleavage of such eggs is abnormal and they degenerate in a short time, like the eggs laid by unfertilized females. An analysis of the destiny of sperms after copulation was carried out in strains from the Leghorn Aquarium, which were bred in the laboratory under constant environmental conditions, in order to know how such degenerating eggs originate.

JÄGERSTEN[1] showed that internal fertilization occurs in *D. apatris* after introduction of the sperms through the body wall of the female and called it hypodermic fertilization. JÄGERSTEN, however, did not investigate the behaviour of the sperms after their introduction into the female's body. The histological control of many capsules containing individuals in various stages of development showed the passage of the sperms from the male to the female. Such a passage occurs immediately after hatching when the development of the female is not yet completed.

Figure 1 shows the section of a capsule where male and female embryos are copulating and the sperms are passing from the male to the female. The histological examination of two hundred females at different ages showed that sperms, after the coelomic cavity has been reached, group around the gonad. The ovary can be well identified also in very young females soon after copulation has ended (Figure 2). An evaluation of the number of male and female eggs produced by isolated and fertilized females during their whole life cycle showed that a female is able to produce, on the whole, 40–50 eggs as a minimum and 120–130 as a maximum. The number of sperms, on the contrary, ranges between 100 and 80. This is the reason why some normally prolific females lay non-developing eggs at the end of their life. Evidently these females, in spite of the fact that they have finished their sperm supply, carry on the production and laying of eggs that remain unfertilized.

Cytological control shows in fact that the number of sperms is low (Figure 3) in the coelomatic cavities of fe-

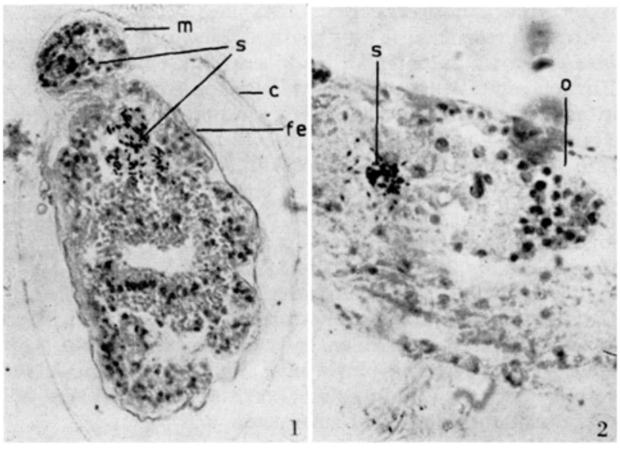

Fig. 1. Fertilization: capsule with the female embryo and the male; fertilization has almost been completed by the male.

Fig. 2. Young female: the sperms received from the male are ready for fertilization.

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[1] G. JÄGERSTEN, Zool. Bidr. Uppsala *22*, 61 (1943).

males which have laid many fertilized eggs. The sperms cannot be found at all in some old females (Figure 4).

De Beauchamp[2] showed in 1910 that *D. conklini* (which is now identified with *D. apatris*) can reproduce by parthenogenesis; Hartmann[3] also obtained the same results in this species. No parthenogenetic development has been obtained in our strains in hundreds of essays consisting in the early removal of the male partner.

It appears, however, that a few of the numerous unfertilized eggs that are laid by old female individuals have a chance to develop parthenogenetically because they have a genetic tendency to parthenogenetic reproduction. It is therefore reasonable to assume that parthenogenetic strains can originate from such eggs laid by old females.

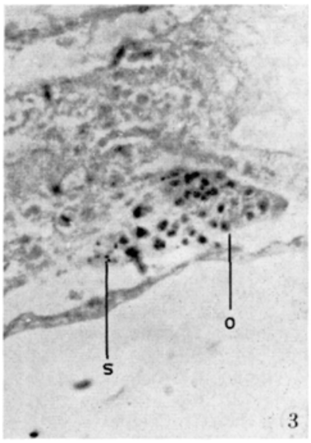

Fig. 3. Adult female: only a few sperms are left.

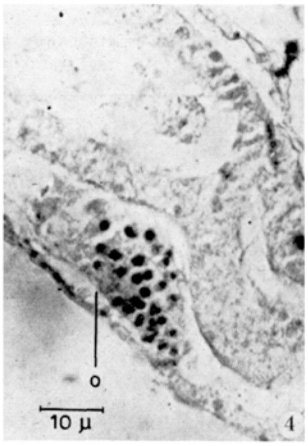

Fig. 4. Old female: all the sperms were utilized.
c, capsule; fe, female embryo; m, male; o, ovary; s, sperms.

Riassunto. La femmina di *D. apatris* è fecondata con un numero di spermi spesso insufficiente per tutte le uova che può produrre durante la sua esistenza, per cui le femmine vecchie depongono uova non fecondate che degenerano a stadi di sviluppo diversi. Questa può essere la causa dell'insorgenza di ceppi partenogenetici.

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Istituto di Zoologia dell'Università di Modena (Italy), January 15, 1965.

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Membrane Constants and Sodium Conductance of a Single Muscle Fibre[1]

Previous results[2] support the validity of 'ionic theory' for the muscle fibre of mammals and suggest that the depolarization is due to an increase of sodium permeability.

The aim of the present research was to measure the membrane electrical constants of the single muscle fibre and to calculate its sodium conductance: the latter was achieved with the results previously obtained[3].

Experiments were carried out on the obliquus abdominis internus muscle of the guinea-pig, separated in a thin layer and kept in a Tyrode solution (aerated with O_2 and CO_2) at 38°C. The membrane constants of a single muscle fibre were determined by the method of 'square pulse analysis'[4], with two intracellular glass electrodes: a rectangular current pulse was sent through a single fibre with one microelectrode, while the second, inserted at a distance of 50 μ and later moved to a distance of 500 μ and 250 μ from the first along the major axis of the fibre, recorded the membrane potential changes; a double-beam oscilloscope was used, the membrane potentials being recorded by one trace and the current monitored by the second channel.

An example of records obtained is given in the Figure showing the electrotonic potential changes due to three different current intensities: applying the cable theory (Hodgkin and Rushton[5]) to the experimental data, the potential changes, E, are related to the steady current, I, by the following equation:

$$E = \frac{1}{2} I \sqrt{r_m r_i} \exp\left[-\frac{x}{\sqrt{r_m/r_i}}\right]$$

where x = microelectrode separation; r_m = transverse resistance of unit length of membrane; r_i = internal longitudinal resistance per unit length of fibre.

Plotting $^1/_2 I \sqrt{r_m r_i}$, on a logarithmic scale, against the interelectrode distance x, the value of $^1/_2 I \sqrt{r_m r_i}$ at the zero distance may be obtained graphically; λ, the space constant, was calculated from the same plot, measuring the distance of the electrotonic potential drop to 1/e; the value of 125 Ω was used for R_i, myoplasm resistance[6]; finally, τ_m, the time constant of the membrane, was obtained from records, measuring the time taken for the potential to rise to 83% of its maximum steady value.

The results are given in the Table, where data obtained for seven different sets of measurements are summarized, each experiment being performed on the same muscle fibre: these data characterize the electrical properties of the resting fibre.

Comparing results from human intercostal muscles[7] and cat tenuissimus[6] it is clear that for the guinea-pig the membrane resistance is lower, while the capacity is the same ($C_m = \tau_m/R_m = 3.8 \pm 2\ \mu F/\text{cm}^2$).

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[1] Partially supported by a grant from the Italian 'Consiglio Nazionale delle Ricerche'.

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[6] I. A. Boyd and A. R. Martin, J. Physiol. *147*, 450 (1959).

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