bactéries, exigentes en facteurs nutritifs contenus dans le sang ou le sérum, cessent de se multiplier et meurent. Seuls un petit nombre de streptocoques moins exigents survivent. Leur multiplication est responsable de l'augmentation de la population à laquelle on assiste durant la seconde période.

Des expériences identiques ont été faites avec d'autres souches de streptocoques du groupe A. Le phénomène d'éclipse n'a été observé qu'avec deux autres souches extremmement virulentes pour la souris (dose léthale voisine de 1 chainette). Nous n'avons jamais observé ce phénomène avec des souches de moindre virulence.

Summary. The streptococci belonging to some strains of group A, when cultivated in small number in an artificial medium, start to multiply only after a hidden period of several hours during which most of them die. This phenomenon is observed only with bacteria obtained from a culture made directly from the heart blood of mice infected with virulent group A streptococci.

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Fine Structure of Chromatic Granules in *Trichomonas vaginalis* Donné

Cytoplasmic structures previously called chromatic granules[1,2], and recently in papers on electron microscopy named according to their position in a cell as paracostal and paraaxostylar bodies, may be found in many flagellates. In my study on the fine structure of *Trichomonas vaginalis*, chromatic granules were encountered frequently. When some modifications in the preparation of the cells for electron microscopy were introduced, these granules showed morphological features so far not described in the literature. Ultrastructure of chromatic granules in *T. vaginalis*, as observed after application of a variety of procedures, will be reported in the present paper.

Materials and methods. The strain of *T. vaginalis* Donné isolated from human vaginal excretion was cultivated axenically, according to methods of JOHNSON et al.[3] and FILADORO and ORSI[4]. Two main procedures were used: (1) Centrifuged cells prefixed in phosphate buffered 6% glutaraldehyde (pH 7.2, 0.1 *M*), postfixed in 2% phosphate buffered OsO_4 (pH 7.2) were embedded in Epon. (2) Preserved cellular monolayers[5] prefixed in 5% s-collidin buffered glutaraldehyde (pH 7.2, 0.1 *M*), postfixed in 4% unbuffered OsO_4 (pH 6.0) were embedded in Epon. Sections of cells fixed with both methods were

[1] C. F. T. MATTERN, B. M. HONIGBERG and W. A. DANIEL, J. Protozool. *14*, 320 (1968).
[2] D. H. WENRICH, J. Morphol. *36*, 119 (1921).
[3] G. JOHNSON, U. M. TRUSSEL and F. JAHN, Science *120*, 126 (1945).
[4] F. FILADORO and N. ORSI, Antibiotics chemotherapy *8*, 561 (1958).
[5] F. FILADORO, Rivista di Biol. *62*, 167 (1969).

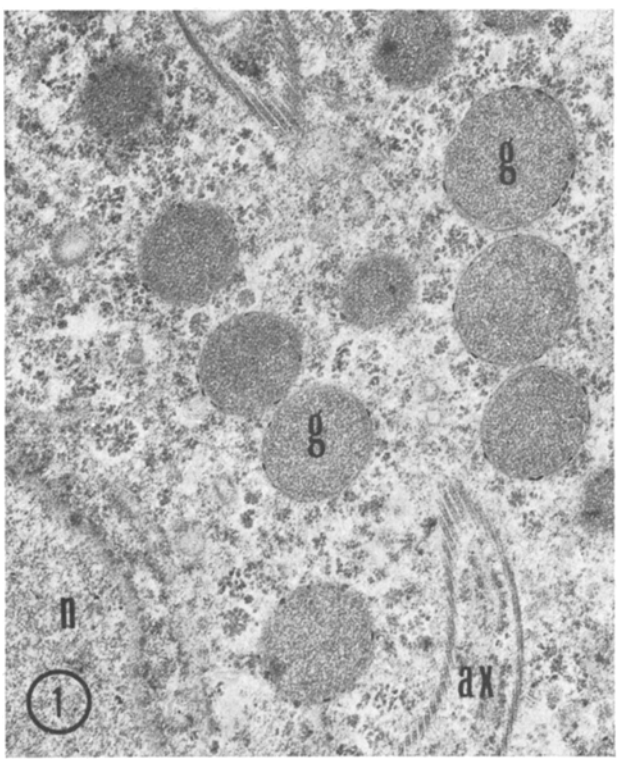

Fig. 1. Chromatic granules (g) in *T. vaginalis* disposed along and around the axostyle (ax). The nucleus is marked (n). The cell was prepared with method 2. × 24,000.

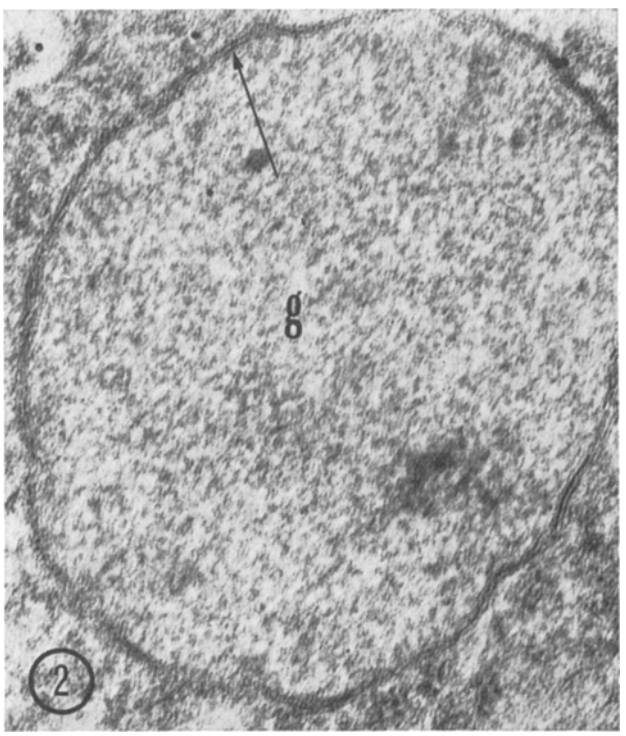

Fig. 2. A chromatic granule (g) of *T. vaginalis* fixed with method 1. No inclusions are seen on the periphery of the granule. An arrow indicates a part of the envelope where a single membrane of the trilaminar type is visible. × 98,000.

stained with lead and/or uranyl acetate. The observations were carried out with Siemens Elmiskop IA electron microscope.

Results and discussion. Chromatic granules in *T. vaginalis* were present predominantly along and around the mastigont system (Figure 1). Their fine structure differed greatly depending on the method of fixation.

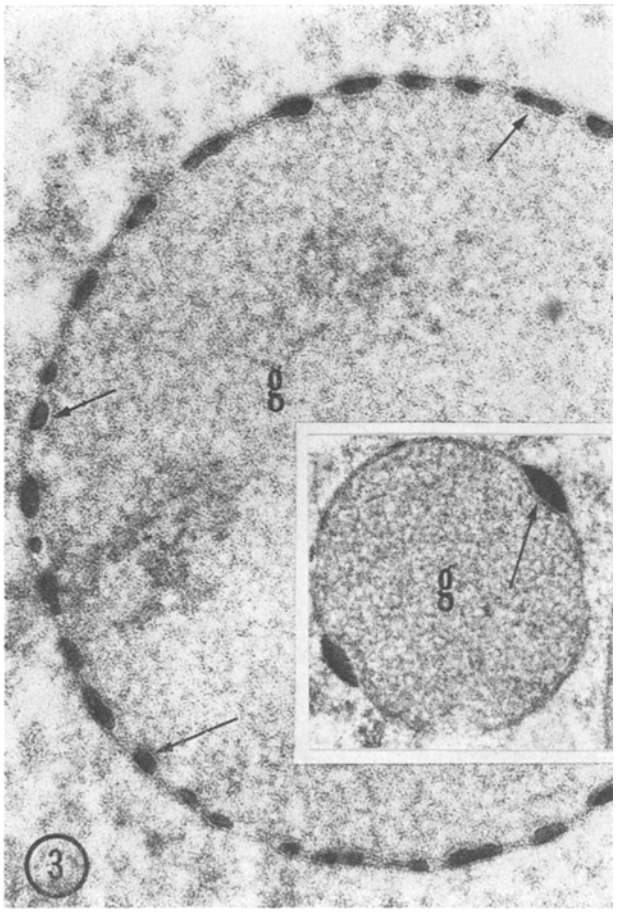

Fig. 3. Higher magnification of a chromatic granule (g) of *T. vaginalis* fixed with method 2. Numerous electron-opaque inclusions are visible in the envelope of the granule. The inclusions surrounded by an optically empty layer 30 Å thick are indicated by arrows. × 98,000. Insert: Higher magnification of a chromatic granule (g) containing only 2 lens-shaped inclusions (arrow) in its envelope. Fixation with method 2. × 96,000.

Chromatic granules fixed with method 1 were limited by a single trilaminar membrane which occasionally had only additional dark and light bands (Figure 2). After application of method 2, lens-shaped inclusions were found along the envelope of chromatic granules (Figures 1 and 3). Between inclusions, the envelope presented the usual aspect of a single trilaminar membrane. Each inclusion was covered on both sides with a 25–30 Å thick optically empty band belonging to the envelope of a granule (Figure 3). Inclusions were either homogenous or contained optically empty core; they were situated regularly but their length varied from one granule to another (compare Figure 3). Their width was more constant and ranged from 60–80 Å. Inclusions were seen in all cells fixed with method 2.

Fixation with 4% unbuffered OsO_4 permitted us to observe the characteristic inclusions in the envelope of chromatic granules, the as yet unknown structural detail. It is possible that higher percentage and lower pH value of osmium tetroxide used in method 2 prevented the extraction of some component responsible for the morphological appearance of inclusions. The reproducibility of findings suggests that inclusions may correspond to structures present in vivo in *T. vaginalis*. The finding of the inclusions does not facilitate the classification of chromatic granules or the understanding of their functional significance. Some ultramorphological similarities between chromatic granules and microbodies[6] may be indicated. Inclusions described in the present paper may correspond to marginal plate or other component of microbodies. So the possibility arises that chromatic granules might be an equivalent of microbodies which were up to now identified in Protozoa on biochemical basis only[6]. To test this assumption, specific cytochemical reactions must be carried out.

Riassunto. Nel presente lavoro viene descritta l'ultrastruttura dei granuli cromatici di *Trichomonas vaginalis* Donné. A seguito di modificazioni alle metodiche della fissazione delle cellule per la microscopia elettronica sono state osservate delle inclusioni nella membrana di questi granuli non descritte in letteratura.

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[6] Z. HRUBAN and M. RECHCIGL JR., in 'Microbodies and related particles', Int. Rev. Cytol. Suppl. 1 (1969).

Induction of Female Flowers on Male Plants of *Cannabis Sativa* L. by 2-Chloroethanephos-phonic Acid

Sex expression in *Cannabis sativa*, a dioecious annual, can be modified by temperature, day length, as well as exogenous application of auxin[1]. The present investigation was undertaken to test whether Ethrel (2-chloroethanephosphonic acid), a recently recommended source of ethylene, could induce femaleness in male plants of *Cannabis sativa*.

Material and method. Seedlings of *C. sativa* were raised in earthen pots. They flowered 8 weeks after germination and their sexes were determined; 60 male plants were selected for treatment and the height and number of vegetative and flowering nodes were recorded for each plant.

Three concentrations (240, 480 and 960 ppm) of Ethrel were applied in one foliar spray (using a hand sprayer) till the point of run-off; triton X-114 at 0.01% was used as wetting agent. 45 male plants received Ethrel; 15 received only triton X-114 (controls). These plants were kept under natural conditions obtaining during January–March, in the departmental botanical garden. The height and flower number were recorded every week following the treatment.