1. Bromine acting for five minutes on the root-tips of *Allium* has a specific effect on the cell nucleus in the resting stage.

2. The effects induced are shown thirty-six hours after treatment by spindle abnormalities in metaphase and anaphase, and result in polyploidy in a large number of cells.

3. Bromine produces chromosome and chromatid fragmentation; the latter may be followed by reunion.

4. The effect of the bromine is cumulative and depends on the time which elapses between treatment and fixation.

5. The cytological effects induced by bromine strongly suggest that it is another specific mutafacient chemical.

J. Churý
Vl. Slouka

Biological Institute,
Medical Faculty,
Masaryk University,
Brno.
Oct. 29.

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Morphological Variability of the Nuclear Substance and Genetic Changes induced by Colchicine in *Escherichia coli*

After the simple and reliable proof was given of a nuclear substance of desoxyribose type in bacteria by Robinow's method, a number of authors followed its physiological changes with regard to the growth-cycle of bacteria[1-3]. We have tried to determine the effects of chemical substances, which in higher organisms act on the nucleus and its division, on the nuclear substance of desoxyribose type and its physiological cycle in bacteria.

We used for our experiments colchicinum crystal, colchicinum amorphum and colchicinum chloroform. The effect of colchicine was controlled by other polyploidy agents such as chloral hydrate and veratrin. The substances were added to broth, to Koser's synthetic medium with glucose, and to solid agar medium in increasing dilutions of 1 : 500 to 1 : 500,000. In the eight strains which we used in our experiments, we studied first thoroughly the normal morphological and biochemical properties. The chromatin structures were stained by Robinow's method.

In 24-hr.-old cultures, we observed splitting of the nuclear substance either into disconnected particles in the cytoplasm or into a number of minute 'nuclei' irregularly distributed within the cell. These forms of nuclear substance were not observed in cultures with chloroform–colchicine and with the control agents. After cultivation for 48 hours, the bacterial cell lengthened, and in bacteria which normally have no more than four nuclei there were six to eight of them. Later, long polynuclear ('polyploidy') cells developed (these forms arise also with chloral hydrate and veratrin treatment), branched long cells and often whole mycelium-like nets.

At the same time we wished to ascertain whether also the biochemical properties of bacteria change with these morphological changes of the nuclear substance of desoxyribose type. Among the eight strains of *Esch. coli* used were three which could not utilize sucrose. During very many cultivations of these strains in media with sucrose, it was not possible to 'adapt' them or to get spontaneously a sucrose-fermenting mutant. By the analysis of cultures with colchicine, we found, in all sucrose-non-fermenting strains, biochemical mutants which were able to utilize sucrose. We obtained such mutants after cultivation for 24 hours, that is, exactly at the time when the disturbance of the nuclear substance and the minute 'nuclei' could be observed. We detected the mutants on the solidified Koser's medium with sucrose as the sole source of carbon, and added as an indicator bromthymol-blue. On this medium sucrose-fermenting bacteria become yellow colonies of normal size. The adult colonies, which should possess the newly acquired biochemical properties, were then examined in order to determine whether they were real mutants and whether their new characteristics were stable and hereditary.

These experiments thus show that the action of colchicine not only induces morphological changes of the nuclear material of a desoxyribose type in *Esch. coli*, but also simultaneously enables us to detect biochemical mutants. Detailed descriptions of our results will be given elsewhere[4].

J. Šterzl

Institute for Medical Microbiology
and Immunology,
Charles University,
Praha II.

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Recovery and Culture of Tubal Mouse Ova

Fertilized mouse ova have been cultivated *in vitro*, and their development filmed, by Friedrich-Freksa and Kuhl[1], who used as medium a clot of guinea pig plasma and mouse embryo extract containing segments of Fallopian tube[2]. Like Chang[3], I have been working on ovum culture with a view to transplantation of ova. Chang has used rabbits, with serum as a culture medium; I have chosen to use mice, as more readily available, and because (like most domestic animals) they have naked eggs. Seeking a medium readily prepared in large quantities, I have tried saline hen-egg extracts.

The procedure adopted revealed an unanticipated difference between the viabilities of two-cell and later tubal stages; eight-cell ova survived and developed in culture, whereas two-cell ova did not.

Saline (NaCl, 880; KCl, 30; $CaCl_2$, 25; $MgCl_2$, 5; glucose, 108 mgm. per cent; and NaH_2PO_4, 10 mgm. per cent), with 8–9 c.c. of thin egg-white added per 100 c.c., was used as a clear fluid for recovery of ova. This fluid (*p*H 7·5–7·8) was stored in the refrigerator and used up to fourteen days after preparation.