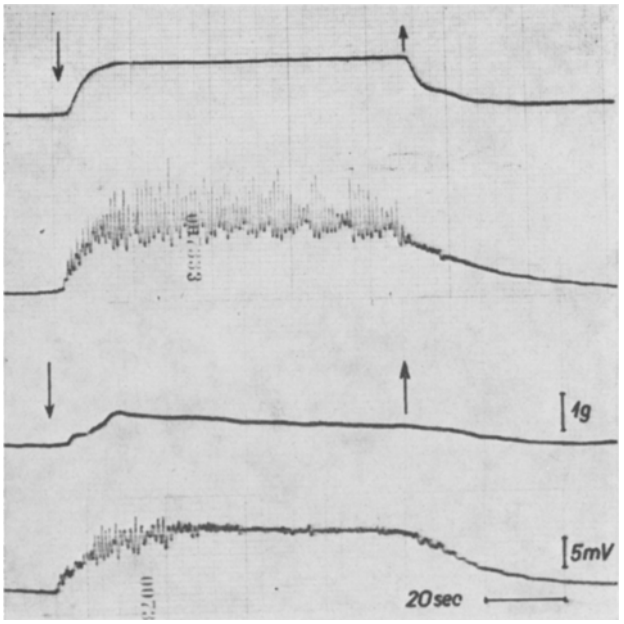


Fig. 1. The mechanical and electrical activities of taenia coli evoked by acetylcholine ($1 \times 10^{-6}$ g/ml) recorded by sucrose-gap method. Top tracing was taken before and bottom tracing during papaverine ($1 \times 10^{-5}$ g/ml) perfusion.

spike frequency induced by acetylcholine was $76.0 \pm 5.1$ per min, and was decreased by papaverine to $67.0 \pm 15.8\%$. Despite the presence of acetylcholine in the papaverine-treated preparation, the spikes and contractions stopped before the acetylcholine was washed out (Figure 1).

The mechanical responses elicited by $BaCl_2$ after papaverine pretreatment were inhibited to $69.5 \pm 12.0\%$. The average spike frequency induced by $BaCl_2$ was $85.3 \pm 5.6$ per min. Papaverine only slightly decreased ($82.0 \pm 14.2\%$) this spike frequency. The results are summarized in Figure 2.

Papaverine was used at a dose level that did not abolish the spontaneous (mechanical and electrical) activities. Shortening of the action of acetylcholine, as opposed to an unchanged duration of the action of $BaCl_2$, after papaverine, agrees with the observation that the responses to acetylcholine were depressed to a larger extent. Papaverine antagonism was maximal against the effects of

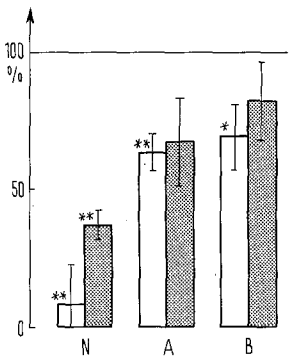


Fig. 2. The comparison of inhibitory action of papaverine on the contractures (open bars) and the spike frequencies (shaded bars) evoked by nicotine (N), acetylcholine (A) and $BaCl_2$ (B). The respective values from 6–10 experiments (mean $\pm$ S.E.M.) were significantly different from controls (100% effect) by $p < 0.05$ (*) or $p < 0.01$ (**).

nicotine. In most cases, the stimulating effect of nicotine was blocked completely. The height of the contractions was always significantly lowered. The increase of spike frequency was significantly depressed by papaverine only in case of nicotine.

Papaverine, in small doses, thus possessed a distinct antinicotinic effect. Since the mechanical responses to acetylcholine and $BaCl_2$ were also, though less significantly inhibited, papaverine may also exert a direct inhibitory effect on smooth muscle cells.

HIRTZ[1] concluded that a part of the action of papaverine involves parasympathetic nerve endings in the intestinal smooth muscle. Our results confirm the antinicotinic properties of papaverine[2], in the action of papaverine on the taenia coli of the guinea-pig.

*Zusammenfassung.* Mit Hilfe der Saccharosetrennwand-Methode wird nachgewiesen, dass Papaverin antinikotinisch an der glatten Muskulatur (Taenia coli) des Meerschweinchens wirkt.

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## In vitro Induced Disappearance of Nucleoli in Cells Treated with LSD-25

There are some drugs influencing the phase-contrast picture of nucleoli in living cells, e.g. adenosine[1], arabinosylcytosine[2], ethionine[3], and quinacrine[4]. The drugs induce the central light zone in the nucleolus followed by the nucleolar fragmentation. The ultrastructural picture of the deeply transformed nucleolus commonly corresponds to the segregation of the organell[5]. However, the nucleoplasm of the cells treated in this way reveals the apparent rough residue of the nucleolar material distinguishable even in the light microscope.

In attempting to enlarge the extension of drugs capable of transforming the nucleolar phase-contrast morphology, influence of lysergic acid diethylamide (LSD-25) on cells of tissue culture was examined. LSD binding to DNA molecules in vitro[6], and the role of the drug in changing the ratio of nucleosides in nervous cells[7], is known well. Also the ability of the drug to make the chromosomal abnormalities in mitotic[8] and meiotic[9] cells was studied as a possible cause of the observed teratogenic[10] and mutagenic[11] effect of LSD, though some newer papers support the negative results[12–15]. The attributes of the drug including the inhibitory effect in mitotic division[16],

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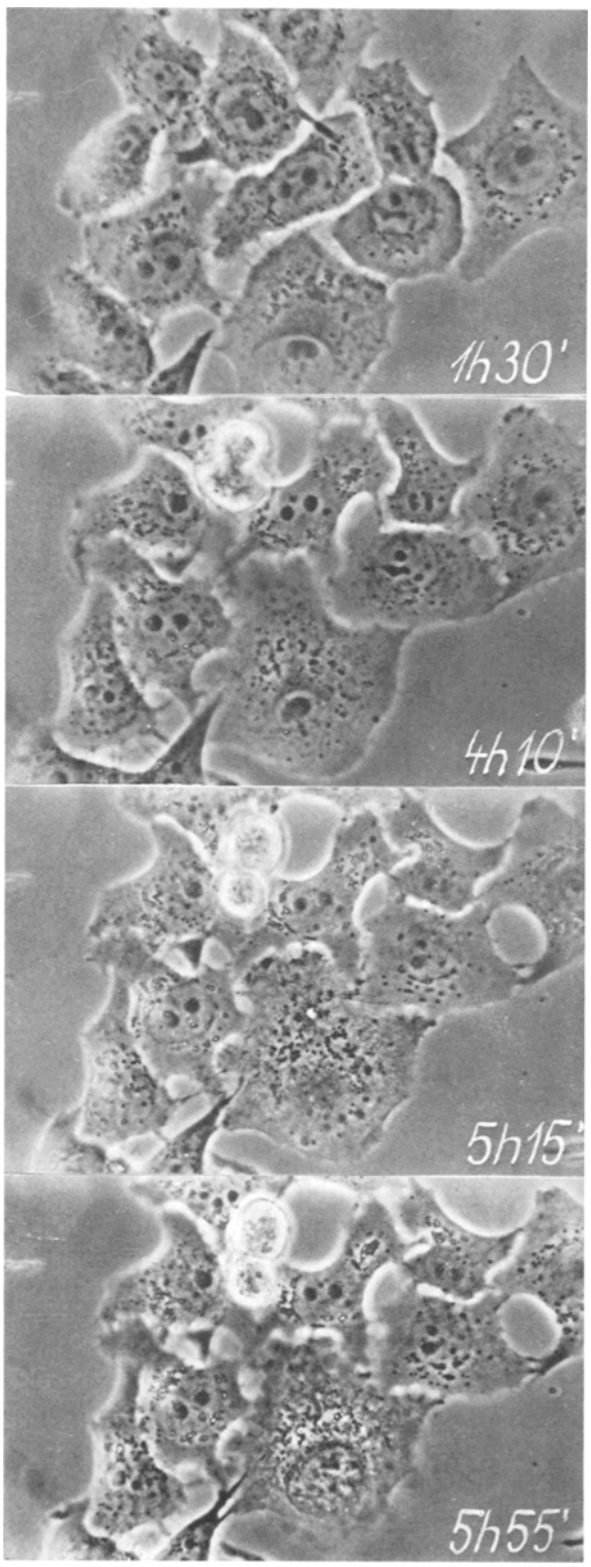
provide sufficient reason for the search of its influences on the structure and behaviour of nucleoli which express in a high degree the functioning of the chromosomal template.




HEp cells 24 h. LSD 100 μg/ml phase-contrast. Time of the cultivation in medium with LSD is signed at the corners of the pictures. × 700.

HEp cells obtained from Dr. Činátl's laboratory (VÚHEM, Prague) were precultured for 24 h in 5 cm Petri dishes on 34 mm round cover slides in synthetic medium Úsol (Prague) with 15% of inactivated calf serum, 1.2% of 0.001% phenol red in bicarbonate buffer, and with antibiotics (200 IU of penicilline and 100 μg of streptomycine per 1 ml of medium). The same mixture was used for the cultivation of cells on cover slides in chambers under the microscope. The concentration of LSD was 25,50 and 100 μg of the drug per 1 ml. The cells were observed in 38 °C by Meopta D microscope with Zeiss phase-contrast condenser, objective 40 and projective 4:1 in green light of the filter VG-4. The time-lapse documentation was provided on 36 mm negative film ORWO NP 15 by camera Praktina.

Nucleoli of cells exposed to LSD changed their shapes and sizes, and some nucleoli disappeared. The process of diffuse disintegration and disappearance of nucleolus, followed by a reconstruction of the nucleus, is represented by the series of photographs. The efficient concentration of LSD was 100 μg/ml.

The other cellular changes were related to increase of the cytoplasmic volume (probably because of cellular flattening), and to distinct separation of granuloplasm and hyaloplasm in some cells. Somewhere, the nuclei move to the peripheral parts of the cytoplasm so that the monolayer forms large cytoplasmic areas. The fusion of the extended cytoplasms is not excluded.

The observed disappearance of nucleoli is not easily explicable. The physiological total disintegration of nucleoli occurs during prophase though not totally regular[17]. The experimental total disintegration of nucleoli was observed by Stockinger[18] in anisotonic media. The process was found to be reversible and repeatable several times. The total diffuse disintegration of nucleoli, interpreted by the model proposed for the nucleolus previously[19] would mean either the sudden large decrease of the synthetic activity of the nucleolar template, or a failure in joining the nucleolar components into the multiple ribonucleoprotein complex structure. As only a small part of the cell population reacts in this way and suffers the disappearance of nucleoli after LSD, the possibility is not excluded that the phenomenon is a function of time, i.e. of the actual position of the cell in its life-cycle.

*Zusammenfassung.* Nachweis, dass LSD Konzentration von 100 μg/ml in der Gewebekultur zur Verminderung der optischen Dichte oder zum Verschwinden der Nucleoli führen kann.

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