Scintillationsmedium aufgeschlämmt und ausgemessen. Die Bestimmung der Mitoseindices erfolgte an feulgengefärbten Quetschpräparaten, desgleichen die mikroautoradiographische Analyse der DNS-Synthesephase, wozu die Strippingfilm-Methode verwendet wurde[6].

Anzahl der Mitosekerne nach 24stündiger Na_2SO_3-Behandlung

Na_2SO_3 (ppm)	Mitosekerne (‰)
0	107 ± 4[a]
10	103 ± 7[a]
100	64 ± 13
1000	45 ± 5

[a] Differenz nicht signifikant.

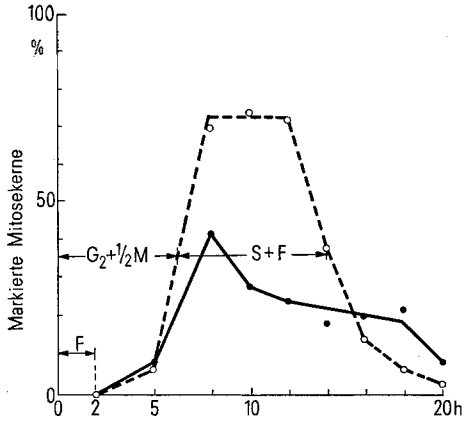

Fig. 2. Mikroautoradiographische Analyse der DNA-Synthesephase im Wurzelmeristem nach 2stündiger Na_2SO_3- und ^3H-Thymidinfütterung (1 µC/ml). ○, Kontrolle; ●, 100 ppm Na_2SO_3.

Unter dem Einfluss von 100 ppm Na_2SO_3 in der Lösung sinkt die Thymidinfixierungsrate auf etwa die Hälfte (Figur 1). Ebenso lässt sich unter der Einwirkung verschiedener Na_2SO_3-Konzentrationen eine Abnahme der Mitosekerne feststellen (Tabelle), wobei die relativen Längen der einzelnen Mitosephasen unverändert bleiben. Die mikroautoradiographische Analyse ergab einerseits eine vergleichbare Länge der DNS-Synthesephase in der Kontrolle mit den Literaturangaben[7], anderseits eine geringfügige Verlängerung unter dem Einfluss von 100 ppm Na_2SO_3 (Figur 2). Zusätzlich deutet der Kurvenverlauf der sulfitbehandelten Wurzeln auf eine Limitierung der DNS-Synthese.

Zur Erklärung dieser Befunde können die an Mikroorganismen und an in-vitro Versuchen gewonnenen Angaben herangezogen werden[4,8,9]. Sie haben ergeben, dass Sulfit unter «physiologischen» Bedingungen mit Cytosin zu einem unverwertbaren Derivat reagiert. Eine Elimination des endogenen Cytosinpools durch Sulfit in den Wurzelzellen wäre denkbar und müsste zur dargestellten Hemmung der DNS-Synthese führen.

Summary. Sulfite inhibits the DNA-synthesis in the root meristem from *Vicia faba* L.

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Nucleolar Behaviour in Lemon Fruit Explants (*Citrus Limon* L.) Incubated in Stabilized and Unstabilized Liquid Paraffin

Medicinal grade liquid paraffin may but need not necessarily contain added stabilizing agents such as tocopherol (vitamin E) or butylated hydroxytoluene (BHT)[1–3]. Although medicinal grade liquid paraffin is easily obtained from a pharmacy, it was not employed in previous investigations concerning endogenous nucleolar behaviour[4,5] or endogenous starch production[6] because no specific information was given regarding the presence or absence of added stabilizing agents. Since BHT is frequently used as a stabilizing agent in edible fats and oils[7–10] including medicinal grade liquid paraffin[1,3], it was considered to be of interest to examine nucleolar behaviour in lemon fruit explants incubated in stabilized and unstabilized liquid paraffin.

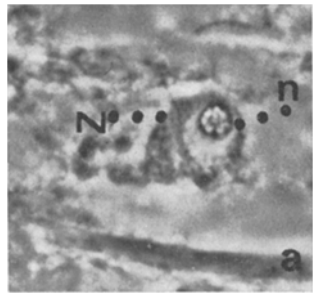
Materials and methods. Entire juice vesicles (sac plus stalk) were excised from surface-sterilized mature lemon fruits (*Citrus limon* L.) as described previously[5] and completely submerged in liquid paraffin with and without 10 ppm of BHT[11] in 'Pyrex' Petri dishes[12] immediately upon removal from the fruit. The stalks were severed from the sacs while beneath the liquid paraffin and then completely immersed in stabilized and unstabilized liquid paraffin in a different set of 'Pyrex' Petri dishes. The Petri dishes in all treatments were sealed with 'Parafilm' and placed in the dark for 48 h at 25–27°C. All Petri dishes and unstabilized liquid paraffin were sterilized by autoclaving whereas stabilized liquid paraffin was used in an unsterilized condition in order to avoid any possible loss of reduced BHT as a result of oxydation during autoclaving. Unsterilized unstabilized liquid paraffin was also employed for comparison with the unsterilized BHT-containing incubation media.

After 48 h, the stalks were removed from all treatments and blotted well with filter paper to remove the excess liquid paraffin clinging to the tissue surface and then placed in Lillie's AAF solution[13] or in a solution of 10% neutral formalin in absolute ethanol (v/v) for 18–24 h in the cold. The fixed explants were dehydrated in absolute isopropanol and unstained paraffin sections were prepared as described previously[5]. Unstained paraffin sections of freshly excised stalks with and without briefly dipping them in unstabilized liquid paraffin before placing them in the fixing solutions served as physiological and

cytological controls. All observations were made with positive phase-contrast microscopy.

Results and discussion. The control tissue showed the typical spindle-shaped nuclei with the spherical-shaped homogeneous-appearing nucleoli as observed previously[4,5]. Marked nucleolar enlargement as well as the formation of highly refractile nucleolar inclusions observed in previous investigations[3,4] were also evident here in the explants incubated in the stabilized and unstabilized liquid paraffin (Figure). There was no evidence of microbial contamination in any of the explants incubated in the unsterilized stabilized and unstabilized liquid paraffin (see also reference No. 6).

The levels of BHT employed as an antioxidant in foodstuffs (ca. 0.01%) show no detectable toxic effects in animals[7–9]. Impairment of growth and of phospholipid synthesis and liver damage have been found to occur in rats at higher levels of BHT (ca. 0.2%)[8,9]. The 10 ppm of BHT employed in the liquid paraffin used in this investigation is the maximum allowable concentration listed in the British Pharmacopoeia[1] for medicinal grade liquid paraffin and is well below the concentrations of BHT considered to be non-toxic and toxic in animal feeding experiments[7–9]. The results of this investigation show that nucleolar enlargement and the formation of refractile nucleolar inclusions were not inhibited by the 10 ppm of BHT in the liquid paraffin used here. In addition, endogenous starch formation was also evident in the sac cells of juice vesicles incubated in the stabilized and unstabilized liquid paraffin (unpublished observation; see also reference No. 6).



48-h-old explant incubated in stabilized light grade liquid paraffin showing an enlarged nucleolus with prominent refractile inclusions. ×1600. N, nucleus; n, nucleolus.

It is not known whether the lemon fruit explants would have responded differently to higher concentrations of the antioxidant BHT. However, the ability of non-growing lemon fruit explants to manifest certain endogenous cytological and physiological phenomena which are also found in growing explants by incubating them in non-aqueous oily media[4–6] may provide a tissue system for examining the effects of fat-soluble antioxidants and their oxidation products on these endogenous cytological and physiological phenomena.

Sumario. Explantas de fruta de limón manifestan semejanto nucleolar comportamiento, cuanádo están incubadas en parafina liquida con, y sin auto-oxidantes.

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Synaptic Boutons in the Hippocampus: Changes are Produced by Age and Experience

For many years researchers investigating the fine structure of the central nervous system have been concerned with the mechanisms by which postnatal structural changes in neurons occur morphologically. It has been suggested that morphological changes in the cortex of the rat after day 20 can be accounted for by means of increased dendritic branching[1]. Furthermore[2,3], researchers have reported that evironmental manipulations alter the fine structure of the dendritic plexus. Based upon these data it seems reasonable to conclude that a possible morphologic site of change exists in the dendritic arborizations of neurons in the central nervous system. Such changes would be expressed by arborization changes within not only the dendritic plexuses but also the axonal endings. Specifically these would include increased dendritic spines, increased axonal arborizations, greater numbers of dendrites, etc. Suggestions such as these have been offered by several investigators[4–6]. Taken together, these morphological alterations suggest that the most sensitive measure of change in dendro-axonal relationships is the number of synaptic boutons

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