frequencies, for the power density of 10–15 mW/cm² has resulted in varying degree of testicular damage such as edema, fibrosis and coagulation necrosis of seminiferous tubules in humans and animals.

This study indicates that nonionizing radiation at 1.7 GHz, 50 mW/cm² for 30–40 min exposure altered spermiogenesis, however, in depth studies should be conducted using the Dominant Lethal Test. At 3.00 GHz, 50 mW/cm² and 20 min exposure the injuries were minimal.

Zusammenfassung. Nachweis histologischer Veränderungen am Hodengewebe 2 Monate alter Schweizer Mäuse nach Mikrowellenbestrahlung. Es ergab sich, dass eine nichtionisierende Bestrahlung bei 1,7 GHz und einer Intensität von 50 mW/cm² während 30–40 min die Samenbildung verändert.

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Bio-Environmental Engineering and Sciences Research Laboratory, Department of Civil Engineering, Howard University, Washington (D.C. 20059, USA), 16 September 1974.

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Studies on the Physiology of Hyacinth Bulbs VII. Root and Bulblet-Like Regenerations from the Ovary Wall of *Hyacinthus orientalis* L.

In horticultural practice, hyacinths are mostly propagated by 'scooping', where the basal plate of bulb is scooped out, or by 'scoring' (cross-cutting), where cuts are made across the base of bulb, each cut being deep enough to cross the basal plate and the growing point. Other methods of propagation of hyacinths are those by 'scaling' and by leaf cuttings[1].

Root and bulblet regeneration of few-month-old hyacinth bulbs[2] and of the perianth of flower buds[3] was found to be dependent on the levels of growth substances added to the MURASHIGE and SKOOG[4] (MS) medium under in vitro conditions. The best combination for bulblet initiation was 1 ppm of α-naphthaleneacetic acid (NAA) and 10 ppm of benzyladenine (BA). For callus formation and root differentiation, NAA was the only necessary hormone.

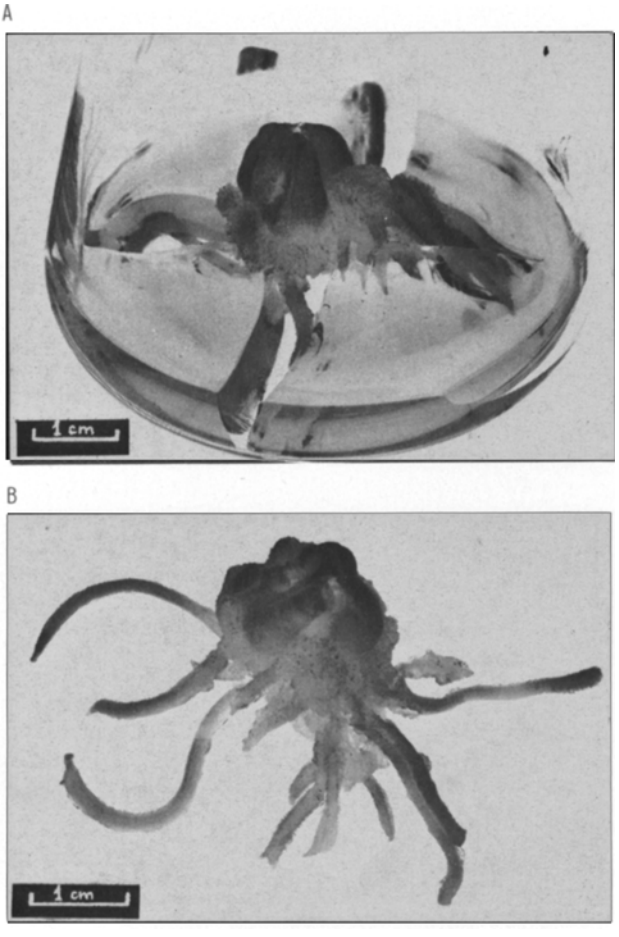

Fig. 1. A 65-(A) and 78-(B)-day-old, in vitro grown ovary of hyacinth on the MS medium containing 1 ppm of NAA: callus and roots formation from the ovary wall can be observed.

The aim of present experiments was to study hormonal regulation in the regeneration processes of excised ovaries of *Hyacinthus orientalis* L. The excised ovaries of hyacinth, cv. Lady Derby, were taken in the spring at the beginning of flowering (April 19, 1974), sterilized with 0.2% solution of mercury chloride for 0.5 h, washed several times with sterilized water. They were planted on MURASHIGE and SKOOG's medium[4] with 1 ppm concentration of auxin (NAA), 10 ppm of cytokinin (BA) or their mixtures. The experiments were conducted in the dark at 25 ± 2°C and 70% humidity. A minimum of 10 ovaries were used for each treatment and the experiments were performed twice.

The type of organ regeneration from the ovary wall of the hyacinth depended on the level of growth substances in the medium. In the MS medium containing 1 ppm of NAA, the ovaries continued their growth and developed into normal-looking capsules, and the callus tissue was found to develop from the wall at base of ovaries. In turn, the callus differentiated into roots (Figure 1). In the MS medium supplemented with 1 ppm of NAA and 10 ppm of BA, the ovaries continued their growth, and, from the ovary wall, the differentiation of small bulblet-like formations took place (Figure 2).

It was found earlier by MAJUMDAR[5] that excised ovaries of *Haworthia turgida* var. *pallidifolia* (Liliaceae), grown in vitro on White's medium containing 1 ppm of indole-3-acetic acid (IAA), 0.5 ppm of kinetin and 20% of coconut water, produced young plants and callus tissue from the ovary wall. The callus was capable of unlimited growth and also produced plantlets with leaves and roots.

[1] A. R. REES, *The Growth of Bulbs* (Academic Press, London and New York 1972).
[2] M. SANIEWSKI, J. NOWAK and R. RUDNICKI, Plant Sci. Lett. *2*, 373 (1974).
[3] M. SANIEWSKI, Bull. Acad. Polon. Sci., Sir. Sci. Biol., in press.
[4] T. MURASHIGE and F. SKOOG, Physiologia plant. *15*, 473 (1962).
[5] S. K. MAJUMDAR, Planta *90*, 212 (1970).

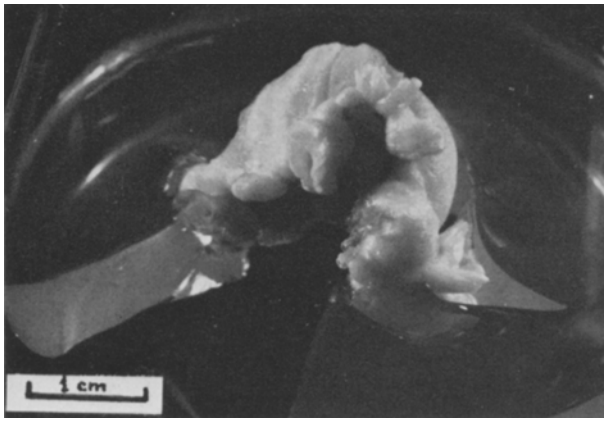

Fig. 2. A 51-day-old, in vitro grown ovary of hyacinth on the MS medium supplemented with 1 ppm of NAA and 10 ppm of BA: the formation of small bulblet-like formations from the ovary wall can be observed.

[6] M. Ziv, R. Konterovitz and A. H. Halevy, Scientia horticult. *1*, 271 (1973).

Organ regeneration of *Alstroemeria* was attempted in various floral parts through tissue culture by Ziv et al.[6]. Callus or root regeneration was obtained from ovaries larger than 5 mm in diameter and branched sections of flower pedicels, but no bud differentiation occurred. All other parts of the inflorescence failed to differentiate. Best callus development was obtained on the cut surface of the ovary planted upright on White medium containing 2.0 ppm kinetin and NAA.

Our study has demonstrated the possibility of differentiation of bulblet-like formations and roots from the ovary wall of *Hyacinthus orientalis* L., and detailed studies of the regeneration processes of this species are in progress.

Zusammenfassung. Isolierte Fruchtknoten von *Hyacinthus orientalis* L. auf Murashige und Skoog-MS-Nährlösung kultiviert, zeigen nach Zugabe von Phytohormonen Ansätze zu Organdifferenzierungen.

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Inhibition of Growth and Respiration of Yeast (*Saccharomyces carlsbergensis*) by Tobacco Smoke Condensate and its Fraction

Extensive experimental work has been conducted in order to develop a quantifiable bioassay for the evaluation of cigarette smoke toxicity[1,2]. The various bioassay procedures that are being used at present involve laborious and expensive methods which at times need to be carried out for a considerable time period in order to obtain any meaningful information[3]. Since yeast cells are relatively easy to grow under various conditions, we have utilized these cells to determine the biological toxicity of the tobacco smoke condensate (TSC) and the water soluble (WS) fraction extracted therefrom. This report describes investigations conducted to determine the effect of (TSC) and its (WS) fraction on the aerobic growth and the respiration of isolated yeast mitochondrial particles.

Materials and methods. All studies were carried out with the yeast, *Saccharomyces carlsbergensis*, grown aerobically in a medium containing 0.2% $MgSO_4$, $7H_2O$, 0.6% $(NH_4)_3PO_4$, 0.5% yeast extract and the growth substrate (2% sodium lactate or 2% glucose), with the final pH adjusted to 5.0 with phosphoric acid.

The growth studies were conducted by growing the organism in 500 ml triple-baffled Bellco Erlenmeyer flasks containing 100 ml of the growth medium. The TSC or the WS fraction was incorporated into the medium to obtain the desired concentration. After 18 to 24 h of growth the cells were harvested by centrifugation, washed once with cold distilled water and the growth was recorded as dry weight.

Mitochondrial particles were prepared from yeast cells grown in lactate medium for 18 h with vigorous aeration. Harvested cells were washed, suspended in a medium containing 0.25 *M* mannitol, 0.02 *M Tris* HCl (pH 7.5), 0.0001 *M* EDTA, 0.2% BSA and broken in a Mini Colloid Mill (Gifford Wood Inc., Hudson. N.Y.). After removal of unbroken cells and cell debris at $1000 \times g$ (5 min), mitochondrial particles were obtained by centrifuging the supernatant at $20{,}000 \times g$ (10 min). The pellet thus obtained was washed once and constituted the mitochondrial preparation. Respiration of cells and the mitocondrial particles was measured polarographically with a Clark-type oxygen electrode and a YSI oxygen monitor (Yellow Springs, Ohio). The proteins were determined by biuret reaction.

The TSC and its WS fractions were prepared by the core services facility of the University of Kentucky Tobacco and Health Research Institute. The U.K. reference cigarettes, (1R1) were maintained at 60% relative humidity and 24 °C temperature. After conditioning for 24 h, the cigarettes were smoked at a rate of 35 ml puff volume for 2 sec/min. Whole smoke was condensed by freezing in glass traps maintained at −60 to −80 °C with dry ice acetone bath. The condensate so trapped was suspended in acetone by sonic vibrations. The WS fraction was extracted from TSC after drying in a water bath at 35 °C under 25″ Hg vacuum, and the resultant condensate was extracted with water and filtered to remove the water insoluble residue.

Results. Aerobic growth of yeast on lactate medium was inhibited by the TSC as well as the WS fractions (Table I).

[1] E. L. Wynder and D. Hoffman, Progr. exp. Tumor Res. *11*, 162 (1969).
[2] F. Homburger, J. natn. Cancer Inst. *48*, 1833 (1972).
[3] B. L. Van Duuren, Cancer Res. *28*, 2357 (1968).

Table I. Effect of tobacco smoke condensate (TSC) and the water soluble (WS) fraction extracted therefrom on the aerobic yeast growth on lactate

Concentration	Inhibition (%) of yeast growth by	
(mg/ml of medium)	Tobacco smoke condensate	WS fraction
0.75	20.0	43.0
1.0	31.0	80.0
2.0	74.2	100.0