

plants grown at 28°C respond much like those grown during the spring and summer. It was consistently found that stomatal aperture was wider in the water control for discs taken from the warm chamber, in accord with the observations of ZELITCH¹ and of DRAKE and SALISBURY². However, the presence of even a micromolar concentration of salt in the bathing solution caused the stomata from the warm chamber to open less than those from the cool chamber, an effect that was observed for all higher salt concentrations as well.

In order to check whether the effect was specific for the salt or due to lowering of the water potential, osmotically comparable concentrations of KCl, NaCl, and fucose were compared. The results (Figure 3) show that the effect is osmotic for all concentrations below 2×10^{-2} eq. l⁻¹. Above this concentration (at least in the experiment of Figure 3) the effect of K⁺ is partially specific,

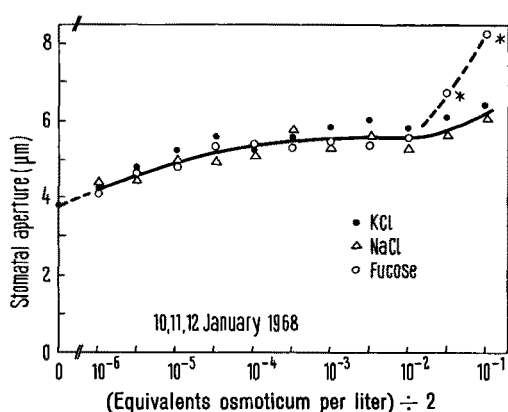


Fig. 3. Comparison of influence of KCl, NaCl, and fucose in the standard assay for stomatal opening. Each point averaged from 3 identical experiments with greenhouse-grown plants. Asterisks indicate wilting of leaf tissue, evidenced only by wrinkles in the replica of epidermal cell surfaces for the first point but for the second point by flaccidity of the leaf disc when it was removed from solution.

as is not surprising in light of the studies of IMAMURA⁵, YAMASHITA⁶, FISCHER^{7,8} and others⁹⁻¹³.

These experiments suggest that the dependence of stomatal opening on previous temperature described by DRAKE and SALISBURY² may be strongly modified by any other variables which influence the water potential of the extracellular solution. If this be true, the adaptive consequences of the temperature effect may be more complex than originally envisioned.

Zusammenfassung. Die Spaltöffnungsweiten in der Epidermis von Blattscheiben aus *Nicotiana tabacum* werden durch osmotisch wirksame Substanzen unterschiedlich beeinflusst, wenn die Pflanzen vorgängig bei verschiedenen Temperaturen aufwachsen. Bei 28°C sind die Spaltöffnungen der Blattstücke in osmotisch wirksamen Lösungen weniger geöffnet als in reinem Wasser, während sich bei einer Temperatur von 18°C die Spaltöffnungsweiten entgegengesetzt verhalten.

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- ¹⁵ The technical assistance of KATHLEEN DUTSON is gratefully acknowledged.

A New Actinomycin-Like Antibiotic Produced by a Mutant Strain of *Streptomyces indicus*

Since the discovery of *Streptomyces antibioticus* in 1941¹, at least a dozen² different *Streptomyces* spp. have been reported for actinomycin-producing cultures. Here we report a pigmented compound, antibiotic MT-10, closely related to actinomycin group of antibiotics. It was isolated from a morphological mutant, strain No. MT-10³, which was obtained through UV-irradiation of *Streptomyces indicus* CHAKRABARTY⁴ (ATCC 25397), a newly described species. It differs from the pink coloured wild type, having yellowish mycelium and spores, and also is capable of diffusing a yellow pigment into the culture media. It also shows antimicrobial activity against bacteria as well as plant and human pathogenic fungi.

The active material was produced in Pridham and Gottlieb's medium (modified) containing (NH₄)₂SO₄, 2.64 g/l; KH₂PO₄, 2.38 g/l; K₂HPO₄, 5.65 g/l; MgSO₄, 7 H₂O, 1.0 g/l; maltose 20.0 g/l, and pH was adjusted to 7.0 before sterilization. The substance was produced in stationary flask cultures at 28°C after 10 days of incubation. The antibiotic was extracted by ether and

purified over alumina column chromatography using ether as the eluting solvent. The active material was obtained as orange yellow crystals from methanol by drying in vacuo.

The homogeneity of the active material was established by paper chromatography and TLC. The compound is readily soluble in ether, acetone, methanol, ethanol, butanol, chloroform, benzene, ethyl acetate, methyl ethyl ketone and acetic acid, but only slightly soluble in carbon tetrachloride and ethylene glycol. It is insoluble in water and petroleum ether. It is decomposed at 204°. Specific rotation is -205-215° (C = 0.25% in ethanol). UV-absorption spectrum shows maxima at 420 and 442 nm. The IR-absorption spectrum shows maxima

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Table I. Antimicrobial spectrum of the pigment antibiotic MT-10

Test organism	Minimum inhibitory concentration (µg/ml)
<i>Staphylococcus aureus</i>	1
<i>Streptococcus pyogenes</i>	2
<i>Bacillus subtilis</i>	0.5
<i>Pseudomonas aeruginosa</i>	50
<i>Candida albicans</i>	90
<i>Candida parapsilopsis</i>	50
<i>Candida tropicalis</i>	80
<i>Trichophyton mentagrophytes</i>	40
<i>Epidermophyton floccosum</i>	35.0
<i>Curvularia lunata</i>	12.5
<i>Alternaria solani</i>	15.0
<i>Fusarium oxysporum</i>	75.0
<i>Helminthosporium oryzae</i>	20.0
<i>Aspergillus niger</i>	125.0
<i>Aspergillus oryzae</i>	35.0

Table II. Physicochemical properties and toxicity of the antibiotic MT-10 and actinomycin D

	Antibiotic MT-10	Actinomycin D
Melting point (°C)	204	240
Specific rotation	$[\alpha]_D^{25} = -205$ to 215° (C = 0.25% in ethanol)	$[\alpha]_D^{25} = -261$ to 268° (C = 0.25% in ethanol)
λ_{max} (nm in ethanol)	420.0 442.0	420.0 440.0
Toxicity in mice LD ₅₀ (mg/kg/body wt.)	0.56	0.76

at 3380–3230 cm⁻¹ indicating the presence of hydroxyl groups. The regions at 1730 cm⁻¹ and 1640 cm⁻¹ are suggestive of the presence of δ -lactone or esters and unsaturated ketone or quinonoid systems respectively. The antibiotic is stable at room temperature. The micro-analytical results shows C — 56.37%, H — 7.40% and N — 10.80%. The mol. wt. is 402 (Rast's method) and the probable molecular formula is suggested as C₁₉H₃₁N₃O₆. The antimicrobial spectrum of the purified substance was determined by cup assay method and its minimum inhibitory concentration is shown in Table I. The toxicity test of the antibiotic was carried out on mice in which LD₅₀ is 560 µg/kg of body weight.

Table III. Comparative in vitro activity of the antibiotic MT-10 and actinomycin D

Test organism	Zone of inhibition in mm Antibiotic MT-10 (100 µg/ml)	Actino- mycin D (100 µg/ml)
<i>Bacillus subtilis</i>	30.5	26.5
<i>Staphylococcus aureus</i>	24.0	20.0
<i>Escherichia coli</i>	—	—
<i>Pseudomonas aeruginosa</i>	22.0	20.5
<i>Candida albicans</i>	22.0	15.0
<i>Candida parapsilopsis</i>	28.5	22.0
<i>Candida tropicalis</i>	19.5	13.5
<i>Saccharomyces cerevisiae</i>	16.0	—
<i>Microsporium canis</i>	13.0	—
<i>Curvularia lunata</i>	22.5	12.5
<i>Alternaria solani</i>	24.0	13.5
<i>Fusarium oxysporum</i>	15.0	—
<i>Aspergillus niger</i>	14.0	—
<i>Aspergillus oryzae</i>	18.5	—

—, indicates absence of activity.

The orange yellow colour of the product, UV- and IR-absorption spectrum, high negative optical rotation, high toxicity and also its solubility indicate its relationship to those of actinomycin group of antibiotics. A comparison was therefore made with actinomycin D (Table II). Regarding solubility in different solvents, both antibiotic MT-10 and actinomycin D behave similarly, except in water where the latter is partially soluble. Comparative assays for antimicrobial activities are given in Table III. It was observed that the inhibitory activity of the antibiotic differs from the actinomycin D to some extent. The activity of the antibiotic MT-10 against *Saccharomyces cerevisiae*, *Microsporium canis*, *Fusarium oxysporum*, *Aspergillus niger* and *Aspergillus oryzae* is significant, whereas with actinomycin D no such activity exists. *E. coli* is, however, resistant to both the antibiotics.

Zusammenfassung. Aus einer Mutante von *Streptomyces indicus* CHAKRABARTY sp. nov. wurde das neue Antibiotikum MT-10 (orange-gelbe Kristalle) isoliert. Die antibakterielle und antifungische Aktivität wurde untersucht und festgestellt, dass MT-10 mit Actinomycin D verwandt ist.

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Chromosome Studies in *Salvia* (Labiatae): West-Himalayan Species

The genus *Salvia* has received attention from numerous authors who have been interested in economic utilization of various taxa, but less attention has been given to naturally occurring species in Himalayan flora. MUKERJEE¹ has reported 24 species from the Indian subcontinent of which 9 species are met in the Western Himalayas. Cytologically, the genus *Salvia* has been fairly worked out. Earlier reports include the work of DELASTING², EPLING et al.³, MEHRA and GILL⁴, SCHEEL⁵, STEWART⁶ and YAKOVLEVA⁷. The present study was undertaken to investigate the cytological nature of the West-Himalayan species of *Salvia*.

Chromosome numbers for 20 Taxa of *Salvia* from the West Himalayas are summarized in the Table. The

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