

The relatively smaller size, the vertical position they occupy in water and the tendency of clumping of *Aedes aegypti* larvae is reported to account for a greater selective disadvantage for this species⁸. This could, however, be well compensated for by the comparatively higher rate of genetic variability they possess. The highest record of mutation load for *Aedes aegypti* is 2.96⁹, while that of *Culex fatigans* is only 0.7¹⁰. Hence, the maintenance of a balanced species frequency is expected in natural populations despite such preferential feeding habits of their predators. If one removes mutation rate and adaptation as an explanation, it is difficult to explain the balanced species polymorphism, despite such selective predation, between *Aedes* and *Culex* in areas of natural populations where they co-exist.

These findings have implications on the biological control programmes of mosquitoes, since many laboratory marker strains stand the danger of being selectively fed upon in field experiments¹¹.

Résumé. Les modes d'alimentation préférées de deux prédateurs, *Culex (Lutzia) raptor* et *Gambusia affinis* sur les larves de certaines espèces de *Culex*, *Anopheles* et

Aedes sont étudiés au point de vue de leur valeur pour la survie dans la nature. Une corrélation entre la variabilité génétique des espèces prédatrices et l'adaptabilité est établie. On propose de modifier pour le contrôle biologique.

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⁸ P. T. RAJASEKHARAN and B. N. CHOWDAIAH, *Oecologia*, Berlin 11, 79 (1972).

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¹⁰ P. T. RAJASEKHARAN and B. N. CHOWDAIAH, *Proc. 56th Ind. Sci. Congr. Abstr.* p. 534 (1972).

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A Note on New Chemical Compounds Isolated from a Fungus Hitherto Unknown

A sturdy millet *Paspalum scrobiculatum* Linn. is cultivated in the Western ghats of Maharashtra in India and also in other areas of the country as a food grain. This grain is consumed as food by the poorest section of the rural population. The plant has been wellknown for its toxicity to animals and humans since many centuries¹. BHIDE and AIMAN² reported the tranquilizing effect on animals of the ethanol extract of the total grain. RAMKRISHNAN and SUNDARAM³ observed the occurrence of *Claviceps* with this millet, which was designated as *Claviceps paspali*, Ster. and Hall. RAMKRISHNAN⁴ found *Sorosporium* and *Puccinia*, which were designated as *Sorosporium paspali*, Mcalp and *Puccinia subshata* Febs. Rutti. The samples of the grain cultivated in Ratnagiri and Kolaba districts of Western ghats of Maharashtra were collected after an initial survey in the field by PENDSE et al.⁵. A number of samples were found to be infested by fungi. A number of fungi were isolated from the ear-heads of the collected samples. The more common and predominantly occurring fungus was further characterized, identified taxonomically as *Phomopsis* sp. and designated as *Phomopsis paspali*. The species differed morphologically and also chemically, producing new mold metabolites, from other species of the same genus found on other host plants.

The fresh unpolished grain with husk intact of *P. scrobiculatum* was collected in November–December from fields, when rainfall in the area was quite normal. It was treated with mercuric chloride (2%) for 0.5 min and washed with distilled water, till it was free from mercuric chloride. The washed grain was incubated on 2% Potato-dextrose-agar solid medium at 24–28°C. The white mycelium of the fungus appeared within 2–3 days and the growth was fairly rapid, with the appearance of pycnidia within 8–10 days. It could also grow equally well on liquid potato-dextrose medium or 4% malt. After 15–21 days, the total culture was extracted with ethanol at room temperature for about 4–6 days and the solvent evaporated at room temperature⁶. The solid dried residue was extracted by solvent, such as ether or chloroform, for 48 to 72 h. After evaporation at room temperature, the

residue showed 2 spots, as I and II, in the thin layer chromatogram (TLC) (silica gel/chloroform-methanol 95:5, vanilline in 50% phosphoric acid as developing agent).

The crude ether extract was then subjected to adsorption chromatography for the separation of the compounds. The procedure adopted was as follows. Chromatography of the ether extract (1.978 g) on silica gel (80 g – Merck 0.05–0.2 mm) with di-isopropyl ether as solvent yielded 1.026 g of the less polar compound I from the earlier fractions. Recrystallization from di-chloromethane/di-isopropyl ether yielded pure I as colourless needles, m.p. 268–269°, $[\alpha]_D^{25} = +63^\circ$ (C = 0.103, methanol). The later fractions gave a mixture of I and II, which was rechromatographed in an analogous manner. Thus, 286 mg of crude compound II was obtained. This gave after recrystallization from dichloromethane/ether, 151 mg of compound II as colourless needles, m.p. 161–165° (Sintering at 147°C) $[\alpha]_D^{25} = +45^\circ$ (C = 0.106, methanol, purity Ca = 98–99%).

From elemental analysis and high resolution mass spectra C₃₀H₃₉N₀₅ (493.2838) was established as a formula for compound I and C₂₈H₃₇N₀₄ (451.2699) for compound II. The NMR-, IR-, and UV-spectra indicated in compound II, the presence of 1 benzyl group, 1 γ -lactam, 2 secondary and 1 tertiary methyl groups 1 exocyclic doublebond and 2 *trans*-double bonds, 2 secondary and 1 tertiary hydroxy group. Compound I is the monoacetate of II and is hydrolysed with K₂CO₃ in methanol/water (25°C, 7 h) in almost quantitative yield to the latter compound. The monoacetate I is transformed by treatment with acetic anhydride/pyridine at 25°C/24 h to a

¹ Kautilya, *Artha-shastra*, Adhi. 4 Adhyaya 3, p. 209.

² N. K. BHIDE and R. AIMAN, *Nature* 183, 1735 (1950).

³ T. S. RAMKRISHNAN and N. V. SUNDARAM, *Sci. Cult.* 16, 5 (1950).

⁴ T. S. RAMKRISHNAN, *Diseases of Millets*, 1st edn. (Indian Council of Agricultural Research, New Delhi 1963), p. 135.

⁵ G. S. PENDSE, U. K. KANITKAR and P. G. DESHMUKH, in preparation.

⁶ N. K. BHIDE, *Br. J. Pharmac.* 18, 7 (1962).

di-acetate, still bearing the free tertiary hydroxy group. From these data, it was concluded that the isolated compounds are new⁷ and closely related to cytochalasine-D⁹⁻¹⁰ (Zygosporine-A)⁸. Therefore, compound I is named Paspaline-P. A number of important biological properties are attributed to cytochalasines¹¹.

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¹¹ S. B. CARTER, Nature 267, (1973).

¹² N. K. BHIDE and G. S. PENDSE, in preparation.

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BHIDE and PENDSE¹² have shown that these compounds, given by i.p. injection in dogs at doses of 1–2 mg/kg are found to produce an effect of tranquility to a certain extent with tremors, ptosis and depression of motor activity. Arousability was present with occasional vomiting or sometimes a bowel motion. There was a good recovery in all these cases. The action of these compounds in mice in doses of 1 mg/kg is problematic. It is not possible to say at present whether the effects observed in dogs can be interpreted as tranquillity or whether they indicate some other kind of activity on CNS, which may be of interest.

Zusammenfassung. Aus dem neuen Mikroorganismus *Phomopsis paspali* wurden zwei neue Metabolite, Paspali-P und dessen Desacetylderivat isoliert, die zur Klasse der Cytochalasine gehören.

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Effect of Cobalt⁶⁰ γ -Radiations on the Biological Activity and Some Physical and Chemical Properties of Oxytetracycline

The possibility of utilizing ionizing radiations for sterilization of antibiotics is receiving considerable attention. Earlier studies^{1,2} have shown that Co⁶⁰ radiations affect some antibiotics by way of changes in physical characteristics and chemical properties, with no change in their potency. In the case of streptomycin, dihydrostreptomycin, pasomycine, streptopenicillin and minomycin, some losses in potency have been known to occur³⁻⁵.

In the present investigation, an attempt has been made to determine the effect of Co⁶⁰ γ -radiations on the biological activity and some physical and chemical properties of oxytetracycline.

Materials and methods. *Staphylococcus aureus* NCTC 6571⁶ and *Escherichia coli* 06⁷ were used. The general procedure of work was adapted from CRUICKSHANK⁸; the details regarding the experimental techniques, viz. the maintenance of organism, incubation temperatures, method of testing of antibiotic activity and the irradiation procedures have already been described elsewhere⁹.

Oxytetracycline was irradiated in the dry state¹⁰ using Co⁶⁰ source at the rate of 0.356 m-rad/h. The duration of exposure was adjusted to utilize doses of 5, 10 and 15 m-rad in 3 different samples each.

Results and discussion. The observed effects of γ -radiations upon the activity of oxytetracycline on *Staph. aureus* and *E. coli* at the stated levels, are summarized in the Table. Whereas no deleterious effect on the activity of the antibiotic was observed at 5 and 10 m-rad doses, the activity against both the organisms was found to be completely lost at 15 m-rad dose. These findings are at variance with the earlier reported observations³. Marked changes in pH and colour were also detected at 15 m-rad dose exposure.

A study of the various samples of oxytetracycline (non-irradiated and the ones irradiated at the 3 stated doses) revealed that the absorption maxima of the untreated compound located at 353 nm was lost and an absorption plateau appeared at about 215–245 nm in the case of the sample irradiated at 15 m-rad dose only. This

Effect of different doses of Co⁶⁰ γ -irradiation upon the activity of oxytetracycline against *Staph. aureus* and *E. coli*, its pH and colour.^a

| Dose (m-rad) | Activity in terms of Zone Diameter (mm)/5 μ g antibiotic disc | | pH | Colour |
|--------------|---|----------------|-----|-----------------|
| | <i>Staph. aureus</i> | <i>E. coli</i> | | |
| 0 | 27 | 23 | 3.0 | Yellowish white |
| 5 | 26 | 24 | 3.0 | Yellowish white |
| 10 | 26 | 23 | 3.4 | Yellowish white |
| 15 | 0 | 0 | 8.0 | Grey |

^a Results shown are the mean values of 3 replications

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⁶ Obtained from P.G.I. of Medical Research and Education, Chandigarh (India).

⁷ Courtesy W.H.O. International Escherichia Centre, Copenhagen, Denmark.

⁸ R. CRUICKSHANK, *Medical Microbiology*, 11th edn. (The English Language Book Society, E. and S. Livingstone Ltd., Edinburgh 1969), p. 896.

⁹ K. G. GUPTA, K. K. VYAS and N. S. SEKHON, Am. J. Pharmac. Sci., in press.

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