extracted 2-3 times with 5 ml portions of MIBK. The extracts were collected in a 25 ml *volumetric* flask, diluted to the mark with the solvent and the absorbance was measured at 400 nm against the reagent blank prepared under similar conditions.

7 August 1982

1. Lal, K., Singh, J. and Gupta, S. P., *Acta Chim. Sci. Hung.*, 1977, **95**, 233.
2. Beamish, F. E. and Vanloon, J. C., *Recent advances in analytical chemistry of noble metals*, Pergamon, Oxford, 1972.
3. Lal, K. and Malhotra, S. R., *J. Indian Chem. Soc.*, 1980, **57**, 233.

ISOLATES OF *ASPERGILLUS FLAVUS* FROM SORGHUM SEEDS AND AFLATOXIN PRODUCTION

B. BHADRAIAH AND P. RAMARAO*
Department of Botany, SKNR Government College, Jagtial 505327, India.
*Department of Botany, Osmania University, Hyderabad 500 007, India.

AFLATOXINS, secondary metabolites of *A. flavus* are the most potent hepato-carcinogens[1]. However, the growth of *A. flavus* cannot be taken as a certainty for the elaboration of aflatoxins, since some strains of species do not produce them[2-5]. An attempt has been made to study aflatoxin production by six different isolates of *A. flavus*, isolated from sorghum seeds collected from various places of Telangana region of Andhra Pradesh. In addition, the efficacy of some fungicides on aflatoxin production by the toxigenic strain in three different varieties of sorghum seeds have also been studied.

Six different isolates of *A. flavus* were isolated from the seeds of five sorghums, and they were designated as HYF_1 (from fields, Hyderabad Dt.), KRF_2 (from fields, Karimnagar Dt.), KHF_3 (from field, Khammam Dt.), HLB_4 (from local variety (Y-75) seed stored in gunny baggs at Hyderabad), HHB_5 (from CSH-5 seeds stored in gunny bags at Hyderabad) and KHP_6 (from stored in underground pit at Khammam Dt.). The isolates maintained on potato sucrose agar slants, were screened for aflatoxin production on Richard's liquid medium following the method described by Subramanyam and Rao[6].

Three important fungicides *viz*,, Bavistin (2-(Methoxy-carbomyl) —Benzimidazole), Thiram (75% Tetramethyl, thiuram disulphide) and Dithane Z-78 (Zinc ethylene bisdithio carbomate) have been tested. Each variety of sorghum seed 25 g was taken in a 250 ml Erlenmeyar flask and autoclaved for 15 minutes. The seeds were then infested with spore suspension of one-week old culture of toxigenic strain of *A. flavus*. After infestation, the seeds were treated with 100 ppm of each fungicide separately and incubated at room temperature (28 ± 2° C). After incubation, the seeds were sprayed with hot chloroform to kill the fungus, dried at 60° C for one hour and analysed for aflatoxin production by the method described by Stoloff *et al.*[7]

It was observed that among six different isolates of *A. flavus*, aflatoxins B_1, B_2 and G_1 were elaborated by KRF_2 isolate, while isolate HLB_4 produced only B_1; others did not produce any series of aflatoxins (table 1). Isolates of *A. flavus* vary in the quality and quantity of aflatoxin production depending upon different factors. Of the total 1390 isolates from different substrates, approximately 60% produced aflatoxins[8]. Strains of aflatoxin producing species of *A. flavus* group of fungi are known to vary greatly in their ability to produce and accumulate compounds of aflatoxin series[9] and some strains of the species do not produce aflatoxins[2,4,5,10]. There are quite a few non-toxigenic strains and morphologically there is no difference between non-toxigenic and toxigenic strains[10,11]. However, the non-toxigenic strains can under some favourable environmental conditions produce toxins. The present work confirms the early observations stated above and it is necessary to find out the factors that regulate the aflatoxin production in various isolates.

TABLE 1

Aflatoxin production by six isolates of A. flavus

A. flavus isolates	Aflatoxin production			
	B_1	B_2	G_1	G_2
HYF_1	—	—	—	—
KRF_2	+	+	+	—
KHF_3	—	—	—	—
HLB_4	+	—	—	—
HHB_5	—	—	—	—
KHP_6	—	—	—	—

All the three fungicides decreased the amount of aflatoxin production (table 2). In the present work, Bavistin proved to be efficient in inhibiting aflatoxin production in sorghum seeds. Experiments have to be done on a large scale and the economies have to be worked out before definite recommendations are made. The CSH-5 seed supported the maximum amounts of aflatoxin B_1 followed by Local (M35-1)

TABLE 2

Effect of fungicides against aflatoxin production in seeds

Fungicides*	Amount of aflatoxin B_1 in ppm		
	CSH-5	Local M35-1	Local Y-75
Bavistin	1.5	0.42	0.06
Thiram	1.9	0.82	0.25
Dithane Z-78	7.5	1.50	0.30
Control	15.0	7.50	0.75

*100 ppm concentration

and Local (Y-75). As such, CSH-5 and Local (M35-1) sorghum seeds are proved to be good substrates for aflatoxin production. CSH-5 and Local (M35-1) seeds possess the higher amounts of carbohydrates and starch[12]. In addition to the environment and the strain variability, carbohydrate content of the seed may enhance the aflatoxin production.

Authors are thankful to Dr Van Egmond, Department of Mycotoxin Research, the Netherlands for generous supply of aflatoxin standards and for the confirmation of aflatoxin production by isolate KRF-7 and to Dr Ramesh Bhat National Institute of Nutrition for permission to carry out a part of aflatoxin analysis. One of the authors (BB) is grateful to CSIR for a fellowship.

9 July 1982

1. Anonymous, *I.A.R.C., monographs*, Vol. **10**, W.H.O. Publication, 1976, 51.
2. Boller, R. A. and Schroeder, H. W., *Cereal Sci. Today*, 1966, **11**, 342.
3. Diener, U. L. and Davis, N. D., *Phytopathology*, 1966, **56**, 1390.
4. Joffe, A. Z., *Nature (London)*, 1969, **221**, 429.
5. Hesseltine, C. W., Sorensen, W. G. and Smith, M., *Mycologia*, 1970, **62**, 123.
6. Subramanyam, P. and Rao, A. S., *Curr. Sci.*, 1974, **43**, 707.
7. Stoloff, L., Nesheim, S., Yin, L., Rodricks, J. V., Stack, M. and Campbell, A. D., *J. Ass. Off. Analyt. Chem.*, 1971, **54**, 91.
8. Goldblatt, L. A., *Food Science and Technology Monograph*, (ed.), Academic Press, New York, London, 1969, 458 pp.
9. Wilson, B. J., *Bacteriol. Rev.*, 1966, **30**, 478.
10. Mehan, V. K. and Chohan, J. S., *Indian. J. Mycol. Pl. Path.*, 1974, **4**, 74.
11. Tulpule, P. G., *Indian J. Med. Res.*, 1969, **57**, 102.
12. Bhadraiah, B., 1981, Ph.D. Thesis, Osmania University Hyderabad.

ISOLATION OF A PHOSPHATE DISSOLVING SOIL ACTINOMYCETE

A. V. RAO, B. VENKATESWARLU AND P. KAUL
Division of Soil-Water-Plant Relationship, Central Arid Zone Research Institute, Jodhpur 342 003, India.

MICROORGANISMS capable of solubilizing mineral phosphates are known to occur in different soils[1,2]. Most of the organisms reported so far include fungi, bacteria and soil yeasts which were found to solubilize unavailable inorganic phosphates both in culture media and soil through the production of organic acids[2,3]. Although it was mentioned that actinomycetes too can solubilize inorganic phosphates[1], yet no detailed information is available. The present paper reports the isolation and characterization of a phosphate solubilizing actinomycete from a desertic soil. Further the phosphorus release as influenced by the period of incubation and temperature is also investigated.

While studying the distribution of phosphorous solubilizing microorganisms in desertic soils, a discrete colony of actionomycete showing a wide clearing zone in plates containing modified Pikovskaya's medium[4] was isolated. This organism was identified as *Streptomyces* sp. from its characteristic sporulation in slide culture. In order to study the phosphorus solubilization by this organism Erlenmeyer flasks of 150 ml capacity containing 50 ml of modified Pikovskaya's medium were inoculated with 1 ml of a 4-day old culture grown in Ken-Knight's broth and incubated at $30 \pm 1°C$. At three days interval of incubation, the pH of the medium and the available phosphorus were determined. In another experiment, the flasks were incubated at different temperatures and the pH and available phosphorus were estimated at the end of 7 days of incubation. Each treatment was replicated thrice. Available phosphorus was analysed by following chlorostannous reduced molybdo-phosphroic acid method[5].

The solubilization of tricalcium phosphate by *Streptomyces* sp. was observed as a clearing zone in the agar medium around the colony. The organism grew very slowly on modified Pikovskaya's medium while on nutrient agar medium it grew faster. The growth was in the form of suspended pellets in liquid medium. The solubilization of tricalcium phosphate was increased with time and reached a maximum by 12 days of incubation (table 1). Ortuno *et al.*[6] also reported the maximum phosphorus-solubilization by 12-15 days of incubation while working with *Aspergillus niger* and *Pseudomonas fluorescence*. The pH of the culture filtrate became acidic indicating the production of organic acids which catalyzed the solubili-