

$\log a_N$ versus σ gave a good straight line¹⁵. Hence, we felt that the a_N value of the radical anion obtained from 3-trifluoromethyl-4-methoxynitrobenzene(III) could furnish evidence for steric enhancement of resonance.

It can easily be seen from the table that for the radical anions (II) generated from di- and tri-substituted nitrobenzenes other than III, the a_N values calculated using the additivity principle¹¹ are in excellent agreement with the observed values. In the case of II obtained from III the calculated a_N value of 9.9 G is significantly lower than the observed value of 10.9 G. This appreciable difference between the calculated and the observed a_N values indicates that the trifluoromethyl group enhances the resonance of the methoxy group with the benzene ring. Due to the presence of the trifluoromethyl group the rotation of the methoxy group about the C—O bond is restricted and, therefore, the methyl group prefers to be oriented *trans* to the trifluoromethyl group. Hence, the probability of its attaining planarity with the benzene ring is enhanced. Thus, the trifluoromethyl group does not sterically inhibit the resonance but actually enhances it.

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EXTRACTIVE SPECTROPHOTOMETRIC DETERMINATION OF PALLADIUM (II) WITH 3,5-DICHLORO-2-HYDROXYACETOPHENONE OXIME

KEEMTI LAL AND MOHAN KATYAL*

Department of Chemistry, D.N. College, Meerut 250 002, India.

*St. Stephen's College, Delhi 110 007, India.

THE reagent 3,5-dichloro-2-hydroxyacetophenone oxime, synthesized earlier,¹ has been used in the extractive spectrophotometric determination of palladium (II). The metal and the oxime react to form a yellow coloured species which is extractable into methylisobutyl ketone (MIBK) in the pH range 2.0–4.5. However, the species is not extracted into the other common organic solvents such as benzene, carbon tetrachloride, chloroform and ethyl acetate. The extract in MIBK shows λ_{\max} at 400 nm and its molar absorptivity is found to be $1.8 \times 10^3 \text{ l.mol}^{-1}.\text{cm}^{-1}$. It obeys Beer's law within the concentration range 11.1–89.0 ppm of Pd(II). The Sandell's sensitivity of the system is $0.0591 \mu\text{g cm}^{-2}$. The reproducibility of the determination is $\pm 1.5\%$. The tolerance was set at the amount needed to cause $\pm 2.0\%$ error in the recovery of the metal. The following ions (in ppm) do not cause interference in the determination of 50 ppm of Pd(II): Cl^- , NO_3^- , CH_3COO^- , SO_4^{2-} (2000 each); Br^- , NO_2^- , Na^+ , K^+ , Ca^{2+} , Sr^{2+} , Ba^{2+} , Cd^{2+} , Al^{3+} (1000 each); I^- , SO_3^{2-} , $\text{C}_2\text{O}_4^{2-}$, Be^{2+} , Mg^{2+} , Zn^{2+} , Hg^{2+} , Cr^{3+} , Zr^{4+} , MO^{6+} , W^{6+} (500 each); tartrate, citrate, Mn^{2+} , Co^{2+} and Ni^{2+} (200 each). Cu^{2+} and V^{5+} interfere seriously. The present method is comparable in sensitivity and selectivity to the well known methods of determining palladium.² It is, however, slightly more sensitive than the method using a similar reagent 3-bromo-2-hydroxy-5-methylacetophenone oxime.³

Reagents

Palladium(II) (0.01 M) solution was prepared by dissolving the requisite amount of palladium chloride in dilute hydrochloric acid. The oxime solution (0.05 M) was prepared in ethanol.

Recommended Procedure

A suitable aliquot of palladium(II) solution was taken and its pH was adjusted between 2 and 4.5. It was transferred to a 100 ml separatory funnel and mixed with a five-fold excess of the oxime solution. The mixture was shaken thoroughly and allowed to stand for 10 minutes. The precipitated species was

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.

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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-carcinogens¹. 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 them²⁻⁵. 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 HYF₁ (from fields, Hyderabad Dt.), KRF₂ (from fields, Karimnagar Dt.), KHF₃ (from field, Khammam Dt.), HLB₄ (from local variety (Y-75) seed stored in gunny bags at Hyderabad), HHB₅ (from CSH-5 seeds stored in gunny bags at Hyderabad) and KHP₆ (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 Rao⁶.

Three important fungicides viz., Bavistin (2-Methoxy-carbomyl) — Benzimidazole, Thiram (75% Tetramethyl, thiuram disulphide) and Dithane Z-78 (Zinc ethylene bisdithio carbamate) have been

tested. Each variety of sorghum seed 25 g was taken in a 250 ml Erlenmeyer 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 Stollhoff *et al.*⁷

It was observed that among six different isolates of *A. flavus*, aflatoxins B₁, B₂ and G₁ were elaborated by KRF₂ isolate, while isolate HLB₄ produced only B₁; 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 aflatoxins⁸. 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 series⁹ 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

<i>A. flavus</i> 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 B₁ followed by Local (M35-1)