1-(butyl carbamoyl) 2-benzimidazol carbamate) at 0.05% at the beginning and in the middle of cropping.

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### PEROXIDASE ISOENZYME PATTERN IN THE LIVING BARK TISSUE AS AN INDEX OF MALE AND FEMALE IDENTITY IN DIOECIOUS BURSERA PENICILLATA (DC) ENGL.

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In an earlier communication it was pointed that in the dioecious Bursera penicillata (DC) Engl., a host of sandal (Santalum album Linn.), the leaf peroxidase isoenzyme pattern in the mature male and female plants as well as young vegetative plants (raised through shoot cuttings) showed characteristic differences which could be of use in determining the male and female identity of the Bursera plants.

The Bursera plants remain leafless from November to March. Fresh foliage is put forth during April and simultaneously tiny white flowers appear which remain on the plant for 6 to 10 days, the pollination taking place within this period. In the female plants, the fruits formed ripen and drop during August. The green leaves continue to remain on the Bursera plants (male and female) upto November. Thus, the Bursera plants remain leafless for nearly five months. Hence the utility of the differences in the leaf peroxidase isoenzyme pattern for determining the male/female identity in these plants is limited only to their leaf-bearing period. It was hence of interest to examine the differences, if any, in the peroxidase isoenzyme patterns in the living bark tissue of these plants and their utility in determining the male/female identity in these plants, as the living bark has no limitation of the availability as in the

case of the leaves. The peroxidase isoenzyme pattern in the living bark tissue of the *Bursera* plants was therefore studied. The results of the study are reported in this note.

For the experiments, twigs were separately taken during (i) the second week of April (flowering period), (ii) the second week of September (corresponding to the post fruit-drop period in the female plant), and (iii) the second week of December (leafless period) from mature Bursera plants (10 male and 10 female) as well as from young Bursera plants (6 male and 6 female) raised through shoot cuttings taken from mature male/female plants as described earlier<sup>1</sup>. In respect of these young plants, which remained vegetative throughout because of their not yet reaching the flowering age (usually 5 years) and in which the leafless period corresponds with that in the mature plants, the twig samples were taken at the same time as in the case of the mature plants. From the twigs, the thin bark layer is peeled off and the outer dead bark portion is scrapped off to get the living bark tissue. This tissue (4 g in each case) was cut into small bits and used for preparing the enzyme extract and for studying the peroxidase isoenzyme pattern by polyacrylamide gel electrophoresis as detailed earlier<sup>1</sup>.

It was observed that the peroxidase isoenzyme pattern in the living bark tissue, while it differed between the male and female plants, remained the same in all the male/female plants during April, September and December, irrespective of the fact whether the plant was the young vegetative plant or mature plant at the flowering or post fruit-drop period, and irrespective of the fact whether the plants were at the leafless period or leaf-bearing period. The peroxidase isoenzyme patterns obtained in respect of the male and female Bursera plants and the  $R_f$  values of the bands are shown in the figure.

It can be seen that characteristic differences occur in the peroxidase isoenzyme pattern in the male and female Bursera plants. While the peroxidase isoenzyme bands with  $R_f$  values 0.34, 0.64 and 0.68 were common both for the male and female plants, that with  $R_f$  value 0.28 remained characteristic of the female plant and that with  $R_f$  value 0.46 remained characteristic of the male plant. It may be pointed out that the bands with the  $R_f$  values 0.34 and 0.46 in the male plant appear faint during the period September to December.

The characteristic differences occurring in the peroxidase isoenzyme pattern in the male and female Bursera plants could be of use, without any limitation of time, in determining the male and female identity in the young Bursera plants raised from seed, and in

| R <sub>f</sub> values |      |  |
|-----------------------|------|--|
| M                     | F    |  |
| • •                   | 0.28 |  |
| 0.34                  | 0.34 |  |
| 0.46                  | • •  |  |
| 0.64                  | 0 64 |  |
| 0.68                  | 0.68 |  |

Figure 1. Peroxidase isoenzyme pattern in the living bark tissue of male (M) and female (F) Bursera penicillata (mature as well as young) during April, September and December.

ambiguous cases in respect of (i) young Bursera plants raised through shoot cuttings and (ii) mature Bursera plants. It may be noted that the Bursera plants are raised both by way of potted seedlings as well as rooted shoot cuttings<sup>2</sup>.

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## A NEW METHOD TO DETECT FUSARIUM SPECIES IN SORGHUM SEEDS

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ALTHOUGH standard blotter method is an universally accepted procedure for routine seed health testing<sup>1</sup>, many important slow growing seed-borne fungicannot be detected precisely due to the overgrowth of saprophytic fungi, thus giving erroneous results<sup>2</sup>. Although Fusarium species can be isolated by Agar

plate method using 0.2% pentachloronitrobenzene (PCNB)<sup>3.4</sup>, such a selective isolation procedure is not available for routine seed health testing. In this paper a novel method of detection of Fusarium spp in sorghum seeds (Sorghum bicolor L.) has been suggested which is a modification of the standard blotter method.

Four hundred seeds of each of the five advanced cultivars namely SPV-104, IS 5675, E-35-1, IS 2042 × IS 225)-2 and Uchv-2 × wa × Nigerian)-2 were surface-sterilised using 1% NaOCl for 3 min, then soaked in different concentrations (10, 25, 50 and 100 ppm) of purified crystalline fusaric acid (5, butyl-picolinic acid obtained from Sigma Chemicals, USA) for 24 hr. Seeds removed after 24 hr were plated on blotters moistened with 0.1% water solution of the sodium salt of 2,4-Dichlorophenoxyacetic acid. The seeds were incubated for seven days as described in ISTA Rule<sup>1</sup>.

The observation showed that in all the samples, Fusarium sp expressed to the maximum extent with the elimination of almost all other seed-borne pathogens, except for some toxin-producing fungi like Aspergillus flavus, A. niger and Penicillium spp (figure 1). Of the four concentrations of fusaric acid used 50 ppm gave maximum expression of Fusarium sp, followed by 25, 10 and 100 ppm. In all the samples incidence of Fusarium increased over the control due to the elimination of other fungal genera which compete with the Fusarium sp. Many earlier workers<sup>5-9</sup>, attributed that fusaric acid has phytotoxic effect and inhibits the seed germination. Induction of wilt symptoms at 10 ppm was reported<sup>10</sup>, due to the toxin phytonivein produced by F. oxysporum f. sp. niveum. In our study, soaking seeds in 50 ppm fusaric acid solution for 24 hr did not affect either the seed germination or seedling vigour.

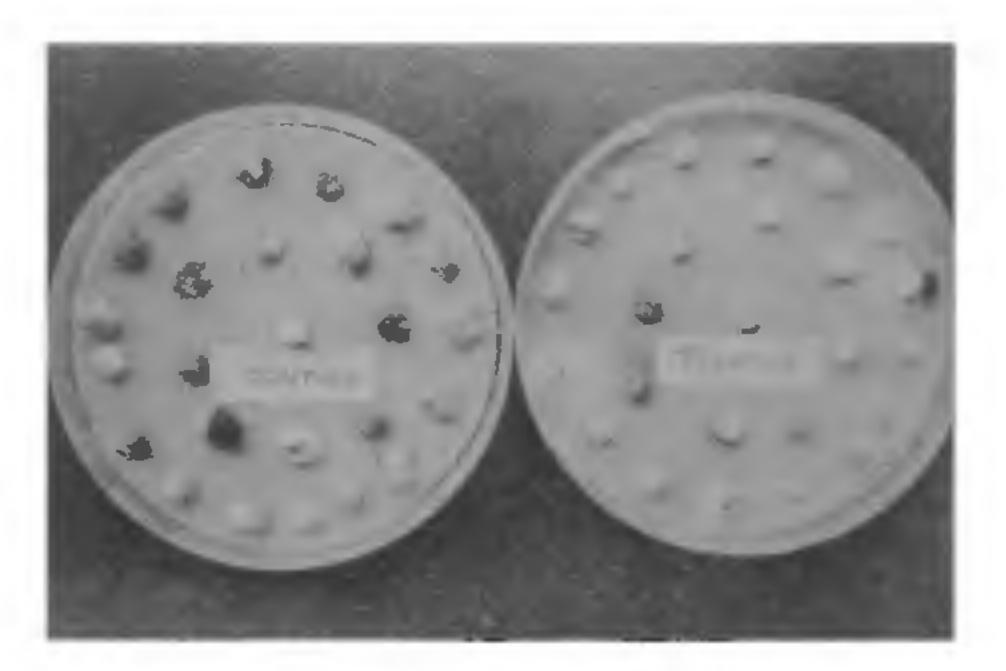


Figure 1. Sorghum seeds treated with fusaric acid showing selective expression of different Fusarium species.