The sorghum genotypes were tall, with sweet stalk, red grains, and take >150 days to mature. In Thailand, hybrid sorghums are predominant over varieties and forage types. In Myanmar, sorghum is grown for food as well as for fodder. Sorghum varieties in Myanmar were similar to those of Thailand and Vietnam. During the survey the crop was in various growth stages from vegetative to physiological maturity or harvestable maturity stages.

### Prevalence of Ergot in Farmers' Fields

The most obvious external symptom of ergot observed on panicles (on nodal tillers or on the main plant) was the honeydew exudation from the infected flowers. Honeydew was either uniformly yellow-brown to pink or superficially dull white. However, no sclerotial stage symptoms were observed.

In Vietnam, ergot incidence was in traces with a severity from 2 to 7%. In Thailand, disease incidence ranged from traces to 80% and severity from 2 to 100% while in Myanmar, disease incidence ranged from traces to 40% and severity from 5 to 80% (Table 1). The samples from Myanmar and Vietnam appear to be *C. sorghi.* Putative *C. africana* types were observed only in the Thailand samples collected from Saraburi, Manavan and Namsuk villages (Saraburi province) and Suphanburi (Suphanburi province). *Claviceps sorghi* was also observed in some samples from Thailand. Reproductive potential of ergot pathogen(s) is an important determining factor, which decides the relative predominance of one species, over the other. Results from molecular analysis are awaited to distinguish species or variability within the species.

**Acknowledgment.** This research project was funded by USDA, USA. The views expressed are not necessarily those of USDA. The cooperation of farmers, non-government organizations (NGOs), staff of agricultural universities, research stations, village councils and Department of Agriculture in Vietnam, Thailand and Myanmar is acknowledged. We thank Suresh Pande, ICRISAT, Patancheru, India and R Bandyopadhyay, IITA, Nigeria for their guidance and help during this survey.

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## Simple Techniques for Production of Secondary Conidia and Ergot Inoculation in Sorghum

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### Introduction

Ergot (sugary disease), caused by several species of *Claviceps* including *C. africana,* is a serious panicle disease in most of the sorghum *(Sorghum hicolor)* producing countries of the world (Bandyopadhyay et al. 1998). Airborne secondary conidia are the primary source of inoculum of *C. africana* (Bandyopadhyay et al. 1998) and are responsible for the rapid spread of the pathogen (Frederickson et al. 1989,1991,1993). Secondary conidia are produced on sterigmata from germinated macroconidia from the honeydew. To date, all infection studies have been conducted with mixed suspensions of macroconidia and secondary conidia, sprayed onto stigmas. However, in nature this does not occur, as the

secondary conidia are not deposited in a suspension on the stigmas, except when carried there by raindrops (Tonapi et al. 2002). In this article, we describe techniques to produce and harvest secondary conidia. and to inoculate stigmas; these techniques mimic natural infection.

## Materials and Methods

**Isolate and sorghum line.** The research was conducted at the School of Agriculture and Horticulture, University of Queensland, Gatton, Australia during 2000/01. The *C. africana* isolate 10765 was maintained and inoculated onto the sorghum male sterile line AQL 33. Secondary conidia were produced by streaking fresh honeydew collected from infected panicles of AQL 33 on 2% water agar and by pouring diluted honeydew suspension on moist soil medium in 9-cm petri dishes and incubated at 20°C in the dark for 36 h.

**Harvesting techniques.** The water agar plates were inverted with the lids partially opened and air moving at 0.2 m $sec^{-1}$ to 3 m $sec^{-1}$ was directed over the secondary conidia. The lid was then closed and the bottom of each petri dish was tapped gently to dislodge the secondary conidia, which were collected inside the lid. In the case of secondary conidia on moist soil, air moving at 0.2 m $sec^{-1}$ to 3 m $sec^{-1}$ was directed over the surface to dislodge the secondary conidia, which were collected on 2% water agar plates held vertically 2 cm downwind of the soil. The efficiency of the harvest was calculated by excising five bits of 1 $cm^2$ sporulating segments of the agar before the secondary conidia were dislodged, shaking them in 10 ml deionized water, and determining the conidial concentration using a hemacytometer. The mean number of conidia per $cm^2$ was then calculated. The mean efficiency of harvest (no. harvested x 100/secondary conidia produced per plate) was then determined.

**Inoculation methods.** Three methods of inoculation (spray, brushing and air movement) with secondary conidia were compared. Twelve panicles of an ergot susceptible sorghum line AQL 33 at 50% flowering were inoculated, and plants were incubated in the greenhouse at 22±3°C for seven days. Each panicle was rated for percentage of infected spikelets.

Spray inoculation: Secondary conidia freshly harvested from water agar plates were suspended in deionized water and the concentration was adjusted to 1 x $10^4$ conidia ml"$^1$. The suspension was sprayed onto the panicles until runoff using a hand sprayer.

Brushing technique: Secondary conidia were collected from the lids of the inverted agar plates with a 1.5 cm wide flat brush. They were then brushed onto the stigmas, which were dry, or which had been moistened by a fine spray of deionized water before inoculation.

Air movement technique: Four blocks of inoculum containing the secondary conidia grown on moist soil medium were placed in the wind tunnel and individual plants with panicles at 50% flowering (whose stigmas had been moistened by a fine spray of deionized water before inoculation) were placed at the same height as the soil, 45 cm downwind of the soil. An inbuilt fan blew air at velocities ranging from 0.2 to 10 m $sec^{-1}$ over the sporulating surfaces towards the flowering panicles. The panicles were then covered with paper bags and incubated in a greenhouse at 22±3°C for seven days.

## Results and Discussion

Of the three inoculation methods, brushing secondary conidia onto dry or moist stigmas was more efficient than spraying them onto stigmas in an aqueous suspension, or using air movement technique (Table 1). Brushing conidia on stigmas produced 70-80% infection compared with 32% with spray and 3% with air movement methods. This difference in infection may be due to the number of secondary conidia that lodged on individual stigmas, rather than the relative efficacy of the method. The results due to air dislodging of secondary conidia through wind tunnel and natural deposition onto the stigmas mimicking the natural infection process resulted in 3.4% infection at wind velocity of 10 m $sec^{-1}$. No infection was observed at wind velocities <10 $msec^{-1}$. The use of air moving across the surface of moist soil on which there is secondary sporulation to dislodge secondary conidia and deposit them on flowering panicles holds potential for infection studies and for the screening of sorghum lines and hybrids for resistance to *C. africana.* Further studies are required to optimize the efficiency of harvest and deposition of conidia on stigmas by manipulating factors such as air speed, relative humidity, temperature and soil moisture.

**Table 1. Inoculation techniques and infection rates using secondary conidia of the sorghum ergot fungus *Claviceps africana.***

| | No. of florets | | Infection |
|---|---|---|---|
| Inoculation technique | Inoculated | Infected | (%) |
| Spray inoculation | 100 | 32 | 32 |
| Brushing onto dry stigmas | 110 | 77 | 70 |
| Brushing onto wet stigmas | 110 | 88 | 80 |
| Air movement | 85 | 4 | 3 |
| Mean | 101.3 | 50.3 | 46.3 |
| SEm± | 5.9 | 19.7 | 17.7 |

**Acknowledgments.** The authors acknowledge the grant of fellowship by J N TATA Endowment, Mumbai, India and provision of facilities by the University of Queensland, Gatton, Queensland, Australia to undertake this research during a postdoctoral visit by the senior author. The isolate of *C. africana* was provided by Sukumar Chakraborty and Birty Koomolong, Cooperative Research Centre (CRC) for Tropical Plant Protection, University of Queensland, St. Lucia, Queensland. Bob Henzell and David Jordan, DPI Hermitage Research Station, Warwick, Queensland provided the sorghum lines AQL 33 used in this study. Special thanks are due to Michael Man and Heidi Robertson, Technical Officers, School of Agriculture and Horticulture, University of Queensland, Gatton for their help and assistance during the course of investigation.

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# Variability and Viability of Sorghum Ergot Sclerotia

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## Introduction

Sorghum ergot pathogen (*Claviceps sorghi* and *C. africana*) infects ovaries that develop into spore bearing masses (sphacelia) in sorghum (*Sorghum bicolor)* panicles. The hard textured sclerotia of *C. africana* rarely protrude more than a few millimeters beyond the glumes while those of *C. sorghi* and *C. sorghicola* may protrude 15-20 mm beyond the glumes. For several ergot pathogens, sclerotia are the resting structures through which they survive in the interval between harvest and the next crop. The sclerotium germinates to produce asci, which produces ascospores that can infect the new crop. How long can these sclerotia remain viable and cause infection? Sangitrao et al. (1997) have reported viability of sclerotia for a maximum of three years. In this article, results on variability and viability of 10-year-old sorghum ergot sclerotia are reported.

## Materials and Methods

Ergot sclerotia collected from sorghum crop during 1992 from Akola (20°70' N and 77° 10' E) in Maharashtra, India were stored under laboratory conditions (25±1"C) at ICRISAT, Partancheru, India. The morphological variability of sclerotia was studied by measuring their size and shape. The viability was tested by pathogenicity tests. To test the pathogenicity, 25 sclerotia of varying morphology were macerated in 30 ml sterilized distilled water using pestle and mortar. The suspension was filtered through sterilized muslin cloth. The filtrate had only mycelial bits and no conidia were seen. The filtrate was made up to 50 ml and was transferred to 100 ml atomizer. The inoculum was sprayed on 10 panicles of sorghum cultivar 296A at 50% stigma emergence stage, using 5 ml panicle$^{-1}$. The inoculated panicles were covered with polythene bags to maintain high relative humidity (≃95%) at 25°C and were placed in the greenhouse (25±2°C) for five days. Before the appearance of honeydew in the panicles, spikelets containing sphacelia