



Evaluation of locally available substrates for mass multiplication of entomopathogenic fungi, *Metarhizium anisopliae* (Metch.) Sorokin

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ABSTRACT

Screening locally available substrates for mass multiplication of *M. anisopliae* such as, broken rice grains, broken maize, broken jowar, broken wheat, and broken ragi grains has showed that, broken rice followed by broken jowar served as the most productive media for conidial production of the fungus, with a yield of 3.45×10^8 and 3.2×10^8 spores per ml, respectively. The next best was broken maize (2.2×10^8 spores per ml), broken wheat (1.94×10^8 spores per ml), and broken ragi (1.8×10^8 spores per ml). Though the broken rice yielded highest spores, broken jowar recorded lowest production cost (8.6Ps/ 1×10^{10} spores), which was followed by broken maize (5.6Ps/ 1×10^{10} spores) proving that the broken jowar was the best substrate for mass multiplication of the fungus.

Key words: *Helicoverpa armigera*, *Metarhizium anisopliae*, Locally available substrates

INTRODUCTION

Other alternatives to chemical pesticides include entomopathogenic fungi, bacteria, viruses, protozoans, nematodes and pheromones that modify the behavior of insects for man's benefit. *Metarhizium anisopliae* (Metsch.) Sorokin (Deutromycotina: Hyptromycetes) is an important fungal pathogen against many insect pests including sucking pests (Sahayaraj and Borgio, 2008). Mass production of the selected bacteria and fungi is a necessary prerequisite for any large-scale field application. However, most of these novel systems have not yet been exploited in agricultural practice on a commercial scale. This is due to the series of sometimes lengthy and sometimes expensive steps, which must be carried out before new pest control systems can be offered to the grower. Hence with a view to find out the highly productive, cost effective and locally available substrates for mass multiplication of *M. anisopliae* the following study was undertaken.

MATERIALS AND METHODS

To evaluate the utility of locally available substrates for mass multiplication of *M. anisopliae* substrates like broken rice grains, broken maize, broken jowar, broken wheat and broken ragi grains were used. The protocol given by Vimala Devi (1994) was followed for this experiment.

Thirty grams of each substrate was taken in 250ml conical flask containing 30 ml distilled water and yeast extract (1%), which was added to all the substrates. After soaking over night, the medium was autoclaved at 15-psi pressure at 121°C for 30 minutes. After cooling, the clump of the substrates was broken and circular agar disks of 5mm diameter were cut from the ten days old fungal culture grown on PDA media containing petriplates. This work was carried out under aseptic conditions using the laminar airflow. The flasks were well agitated for proper distribution of the spores and were incubated at $25 \pm 1^\circ \text{C}$ for 20 days. Four replications were maintained for each

Table 1. Suitability of different substrates for mass multiplication of *Metarhizium anisopliae*

Substrates	No. of spores/ml ($X \times 10^8$)	Total No. of spores produced	Cost incurred for production of 1×10^{10} spores
Broken rice+1% Yeast extract	3.45 ^a	3.45×10^{10}	8.6 Ps.
Broken jowar+1% Yeast extract	3.20 ^a	3.20×10^{10}	5.6 Ps.
Broken maize+1% Yeast extract	2.20 ^b	2.20×10^{10}	6.8 Ps.
Broken wheat+1% Yeast extract	1.94 ^{bc}	1.94×10^{10}	9.2 Ps.
Broken ragi+1% Yeast extract	1.80 ^c	1.80×10^{10}	8.3 Ps.

Means followed by same letter in column are not significantly different by DMRT

substrate and the conidia were harvested by suspending them in 100ml of sterile distilled water containing 0.05 per cent Triton-X-100 (wetting agent). The suspension was filtered through a double-layered muslin cloth and the number of conidia was determined microscopically with a Neubaur improved haemocytometer.

RESULTS AND DISCUSSION

The fungus *M. anisopliae* was mass multiplied on locally available substrates like broken rice, broken jowar, broken maize, broken wheat and broken ragi. The results indicated that the sporulation of the fungus differed significantly among different substrates. Highest sporulation was recorded on broken rice (3.45×10^8 spores per ml) and jowar (3.2×10^8 spores per ml). (Table 1) These two substrates did not differ significantly with respect to sporulation. Broken maize had a sporulation of 2.2×10^8 spores per ml. The lowest sporulation was recorded on broken ragi (1.8×10^8 spores per ml) followed by broken wheat (1.94×10^8 spores per ml), which were on par. The results are presented in table 1. Rice and sorghum contain higher proportion of starch and amylase. Hydrolysis of starch in rice and sorghum resulted in release of glucose and maltose depending on clarification (Preen *et al.*, 1985). Maltose released by the action of starch hydrolysis enzymes present in the fungus induces sporulation (Coudron *et al.*, 1985). Therefore, crushing of seeds was necessary to increase surface area of the substrate for effective action of amylase for hydrolysis of starch.

Based on cost incurred for production of spores, broken jowar recorded lowest cost (8.6Ps/ 1×10^{10} spore), which was followed by broken maize (5.6Ps/ 1×10^{10} spores). Though rice recorded highest spore yield among the substrates, the production cost was higher as compared to jowar and maize.

Quintela (1994) reported conidial production of the fungus *M. anisopliae* on coarse seed was significantly greater than on whole seed. Sorghum and rice seeds served as the most productive media for conidial production of *N. rileyi*, maize was the next best alternative followed by bajra. Similar results were obtained in the present study.

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