

SUMMARY

Title: Investigations on the mechanism of aflatoxin accumulation in cultures of *Aspergillus flavus* and the toxicity of aflatoxin-silica mixtures on rodents.

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Period of Contract: 20 Dec. 1992 to December 1993.

Scientific background and scope objectives of the project:

The high cost of aflatoxins limits the use of aflatoxins for experimental work and routine detection of aflatoxins in contaminated foods. Grated coconut is reported to be an excellent medium for production of aflatoxins under experimental conditions. Various additives are also reported to increase aflatoxin yields in synthetic media. The objectives of the experiments were to identify the optimal conditions for high aflatoxin B₁ yields under experimental conditions, to produce the toxins on a large scale and to study the effect of inhaling of silica gel contaminated with aflatoxins in mice, as humans carrying out research on aflatoxins are exposed to similar conditions.

Experimental method:

Quantities of the cations Zinc, Ferric, Molybdenum and Glycerol already reported to increase aflatoxin production in synthetic media and silica gel were added individually to grated coconut after sterilization at 121 °C for 10 min. in conical flasks. In a second set of experiments the combined effects of the above additives on increased aflatoxin b1 yields was examined.

The flasks were inoculated with spore suspensions of toxigenic *Aspergillus parasiticus* NRRL 2999 in 0.1% Tween 80 and incubated at room temperature in dark for varying periods up to 11 days. The cultures were shaken daily.

Quantitative estimation of aflatoxin produced in the cultures were done by extracting using 85% aqueous acetone and estimating by comparison of fluorescence on TLC. This is a standard method described and widely used.

Results:

Of the different cations tested, Molybdenum did not enhance aflatoxin production. All the other additives enhanced aflatoxin production in grated coconuts to different extent. The combination of additives together with silica gel described in table 1 gave the best yield increase.

Table 1 - Increase of aflatoxin B1 production on addition of silica gel, iron , zinc and glycerol together to coconut cultures.

Additive	Concentration	Aflatoxin B1 (ppm)
Coconut alone	--	206
Coconut + Silica gel	40g/20g of coconut	375
Coconut + Silica gel	40g/20g of coconut	
+ Zinc	12.5 ppm	
+ Iron	25 ppm	
+ Glycerol	1%	412

Conclusions:

Of the different additives tested glycerol and silica gel appears to be the best. The addition of inorganic cations described in literature to increase aflatoxin B1 yields appeared less effective than addition of glycerol or silica gel to the grated coconut.

Papers published:

Presentation at the workshop for grantees held on 29 April, 1994 at NARESA.

Title: Method to enhance yields of aflatoxin in laboratory cultures.