ADVANCES IN METHODOLOGY FOR THE DETECTION OF MYCOTOXINS OCCURRING IN CEREALS AND ANIMAL FEEDS

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INTRODUCTION

Studies at the ADAS Slough Laboratory are aimed at developing methods for a wide range of fungal metabolites to determine which mycotoxins can occur in cereals and animal feedstuffs in the UK. The Poster illustrated some recent developments.

Considerable data on the occurrence of some mycotoxins in the UK have been accumulated over many years. However it is certain that other fungal metabolites also co-occur and probably contribute to the overall effects caused by mycotoxins present in contaminated samples. Reliable and sensitive analytical methods are fundamental to any studies and some developments to improve methods are described

ANALYTICAL CONSIDERATIONS

An analytical procedure typically involves sampling, extraction, clean-up, separation, detection, quantitation and confirmation. Sampling must produce sub-samples for analysis which are representative of the bulk under test. The efficiency with which a solvent mixture extracts mycotoxins present in a sample must be carefully checked especially as extraction of an artificially spiked sample may not give a reliable indication of the extraction of the same mycotoxin formed naturally. Sensitive methods of detection are of restricted value without effective clean-up of solvent extracts. With mouldy samples especially, many co-extracted compounds may still be present in extracts after clean-up and reliable confirmatory methods are necessary positively to identify the compound under study.

CURRENT INVESTIGATIONS

Two improved clean-up procedures are being evaluated. Gel permeation chromatography (GPC) is being studied for use as a multi-mycotoxin clean-up method and a procedure suitable for aflatoxins has been accepted for publication (ref. 1) The method is also being extended to zearalenone, zearalenol, sterigmatocystin, ochratoxins A and B and citrinin.

For specific mycotoxins, immuno affinity columns in which antibodies to the mycotoxin are immobilised on an inert base material and packed into a column

have already been marketed for aflatoxins. An extract containing aflatoxins is passed through the column and aflatoxins bind to the antibody while other material passes through. Subsequently, the antibody protein is denatured and the aflatoxin eluted for analysis. The improvement in clean-up is shown below when either of these two procedures is compared to a commonly used method of analysis, (aflatoxin B₁ at 10 µg/kg and B₂, G₁ and G₂ in an animal feed sample).

Improved sensitivity and selectivity have been obtained for zearalenone and sterigmatocystin using post column derivatisation with aluminium chloride in methanol. This reaction provides additional confirmation for these mycotoxins. Fluorescence and uv absorption are well established as detection methods in mycotoxin analysis but electro-chemical and diode array detectors are alternatives which are being evaluated.

A technique using colour photography of TLC plates under uv irradiation has been developed for recording the spots separated on TLC plates and is proving of considerable value for subsequent re-evaluation of the data as the need arises.

CONCLUSIONS

Considerable improvements in both specific and multi-mycotoxin methods for a wide range of mycotoxins and fungal metabolites have been achieved and reported but further developments are vital for thorough evaluation of the range of mycotoxins occurring in UK grain and animal feedstuffs.

REFERENCE

- 1 M.T. Hetmanski and K.A. Scudamore, A simple quantitative HPLC method for determination of aflatoxins in cereals and animal feedstuffs using Gel Permeation Chromatography clean-up, Food Additives and Contaminants, 1988 (in press).