AUTHOR	SAMARAJEEWA, U
ORIGINAL TITLE	Mycotoxins in coconut based human and animal foodstuffs
DEGREE	PhD
YEAR	1975
INSTITUTE	University of Peradeniya
LOCATION	UP(MED)
MAIN HEADING	TOXINS, BIOLOGICAL
ABSTRACT	An attempt has been made in this study to determine the extent of the aflatoxic problem in Sri Lanka with special reference to coconut products with a view to suggesting preventive and regulatory measures for the control of contamination. The study of the extent of the aflatoxic problem was made by first establishing the most suitable method for the assay of aflatoxins in coconut products. Although several procedures have been described for the assay of aflatoxins in various agricultural products there was no established procedure for the assay of aflatoxins in coconut products. A method based on blending with a mixture of chloroform and water. originally devised for groundnuts, has been used by some workers for coconut products. However no data was available on the comparative suitability of this method as regards its extraction efficiency, interference on thin layer chromatographic estimations and convenience. Therefore the existing methods for the various products were restudied with coconuts to establish the best conditions for each method of extraction. The methods were then compared under optimum conditions. All procedures were found to bring about only partial extraction (about 70percent) of aflatoxins. The 70percent aqueous acetone blending procedure with lead acetate purification was found to be the most suitable on account of convenience and the purity of extracts which enabled titration without a preliminary column purification. Since a constant fraction of aflatoxin present in a given type of coconut product was extracted by a single homogenization by the aqueous acetone procedure, the application of a correction factors for the observed aflatoxin levels has not been described in the literature for assay of aflatoxins. As regards bioassay of extracts, the tadpole bioassay method established earlier for pure aflatoxins was found to be applicable to crude extracts of commercial samples preceded by a short column purification. A field survey was done for the year 1973 to study in detail the extent and

mostly to ignorance and negligence during curing and storage of copra. Education of the copra miller and worker, on the economic and toxicological consequences of contamination is of prime importance in the prevention of contamination. Reorganization of the present inspection network in the mills is suggested as a measure for effective control. The economic loss to the industry due to fungal contamination of copra during storage has been estimated earlier to be around 25percent and such control may therefore minimize these losses. Contaminated coconut oil should be diverted for chemical refining or split into glycerol and fatty acids. The contaminated copra should either be steam treated at high pressure or be rejected totally if the contamination is heavy. Investigation into an outbreak of deaths of young goats of an imported breed at Kottukachchiya (NWP) Government goat breeding centre was shown to be due to aflatoxins. The aflatoxins were detected in the feed samples and in the liver tissues and urine of diseased or dead animals. Strains of Aspergillus flavus were isolated from feed samples and were shown to be toxigenic. Identical clinicle features, postmortem appearances and histopathological lesions were produced by experimental feeding of the goats with artificially infected coconut. The factor that contributed to the toxicity of the industrial feed was found to be an ingredient 'polkudu' added in the preparation of the feed mixtures. As there were significant levels of aflatoxins in local coconut products which could cause toxic and trade problems it became necessary to investigate possible methods of prevention of contamination and detoxification of already contaminated samples. Natural and cheap methods were therefore studied for possible industrial application in decontamination. On account of resistance to fungal growth or aflatoxin accumulation described in the literature for various feed products such as groundnuts, sunflower seeds and soybean varieties, different strains of coconuts were studied for their resistance to fungal growth and aflatoxin accumulation by inoculation of grated coconut with different fungal strains under different conditions of incubation and examined at single or several incubation times. No reproducible results were obtained in any of the experiments. The strains of coconut which showed relative resistance on incubation for a fixed number of days showed equal susceptibility on time course studies. The possible reasons for these discrepancies are discussed. The literature on the resistance of other seed products is also controversial, and it is suggested that time course studies of aflatoxin accumulation may minimize errors in such determinations. The applicability to coconut products of the age old practice of smoking for the preservation of foods was studied by inoculating smoked grated coconut, smoked broth media or broth media containing smoked water for its efficiency in inhibiting aflatoxin accumuulation. The treatment was found to inhibit aflatoxin accumulation to a greater extent than mycelial growth. The moisture content of coconut kernels appeared to be critical as moisture levels above 40percent and

15percent supported aflatoxin accumulation in smoke dried and electrically dried kernels respectively. The inhibition of aflatoxin accumulation by coconut charcoal smoke was found to be apparently due to water soluble fractions in smoke as smoked sterile water or broth were also effective in inhibition of aflatoxin accumulation. As spontaneous loss of aflatoxins was previously observed during storage of contaminated coconut the possible cause of the loss were investigated by storing coconut oil under different laboratory conditions. Of the various natural factors studied (heat, light, enzymic activity) only sunlight was found to be effective. The effect was studied in detail. Detoxification of aflatoxins in coconut oil was brought about by subjecting oil in the form of a thin layer of about 2-4 cm. in thickness under experimental conditions to sunlight. The quality (FFA. coolur) of the treated oil was the same as the untreated oil. Pilot plant experiments with the method are suggested for possible adaptation to industrial decontamination. With solid products, copra and poonac, sunlight was not effective in degrading aflatoxin probably because the sunlight did not penetrate solid particles as effectively as through liquid oil. Some strains of Aspergillus flavus isolated from local copra were found to exhibit intense blue fluorescence on hyflosupercel toxigenicity screening plates. These strains were cultured on grated coconut and the metabolites were extracted. A blue fluorescent compound having different but close Rfs with aflatoxins in several TLC solvent systems was isolated. The compound produced fatty changes in the livers of ducklings after oral dosage. The spectral characteristics and the TLC data suggest it to be a hitherto undescribed factor of possible toxicological interest. The lines on which further research in the field of aflatoxins in coconut products are indicated.