หอสมุดกลาง
จุฬาลงกรณมหาวิทยาลัย

INTRODUCTION

It has been known that the growth of molds on foods is associated with the changes in physical nature of the foods, taste and odor of these food products and appearance of the foods. In addition, certain species of molds can also produce antibiotics such as *Cephalosporium* (Gotshall et al, 1961) and *Penicillium chrysogenum* (Flynn et al, 1964) has a capability to produce penicillin. Recently, another dimension has been added, this is a production of highly toxic metabolites called mycotoxins by many spoilage molds when they grow on foods and feeds. Most of the molds that contaminate on the foods and cereal grains are classified as field, storage and advanced decay fungi (Christensen, 1965). The field fungi invade the grains while these grains are developing on the plants in the fields before harvesting. The principle genera of the field fungi are *Alternaria, Fusarium, Helminthosporium*. The storage fungi invade the cereal grains after harvesting and especially during storage period in the field or the markets.The molds in this group are *Aspergillus* and *Penicillium*. *Fusarium*, a mold in the group of the field fungi, is also found in the advanced decay period of corn along with *Papulospora, Chaetomium* and *Sordaria*.

The toxicity of moldy feeds for livestock has long been recognized (Butler, 1902), and many outbreaks of mycotoxicosis in animals and man have been reported (Sippel et al, 1953; Forgacs and Carll, 1962; Wogan, 1969; Goldblatt, 1969; Shank et al, 1972; Campell and Stoloft, 1974). One important outbreaks

of mycotoxicosis among others is aflatoxicosis among others is aflatoxicosis in which this disease caused many thousands losses of turkey poults and other poultry in England, 1960 (Goldblatt, 1969). The disease in the poultry was characterized by subcutaneous hemorrhages of the legs, and back, body weight lossess and sudden death. At autopsy, the liver was enlarged, pale and firm. It was found that the disease was related to the feeds use of the peanut meal imported from Brazil and certain African countries. Eventually it was learned that the peanut meal was heavily contaminated with mold, *Aspergillus flavus*. This strain of A. *flavus* grew in the peanut meal and produced the metabolites which accumulated and caused the meal to be toxic to the poultry and many animal species. These metabolites have been named aflatoxins even since. A similar disease was soon reported in ducklings and chickens (Asplin and Carnaghan, 1961), in swine (Loosmore and Harding, 1961; Harding et al, 1963), and in young cattle (Loosmore and Markson, 1961). Aflatoxins are detected in four derivatives of B_1, B_2, G_1 and G_2 in variety of foods and foodstuffs. The compounds are coumarin derivatives and aflatoxin B_1 has been shown to be a highly potent hepatocarcinogen in many experimental animals (Goldblatt, 1969). Furthermore, aflatoxins are also toxic to many species of farm animals. The first clinical signs of aflatoxicosis in theseanimals are the loss of appetite and body weight. A few days before death, the animals appear dull, develop ataxia and become recumbent. The most important pathological effect is liver damage including periportal necrosis, hemorrhage and persistence of fat in duckling and extensive bile duct proliferation in some species and fibrosis and venooccussion in bovine are also

seen (Allcroft, 1969). In addition, aflatoxin is suspected of having a role in certain human diseases, and circumstantial evidence has been obtained which suggests that aflatoxin may be a factor in the etiology of Reye's syndrome in Thailand (Bourgeosis et al, 1971a) and in New Zealand (Becroft and Webster, 1972) and of liver cancer in Thailand (Shank et al, 1972c and 1972d) and in Kenya (Peers and Linsell, 1972).

A. flavus is not an only strain that can produce aflatoxins. Aflatoxins were produced by many species of *Aspergillus* and *Penicillium* such as *A. parasiticus*, *A. niger*, *A. wentii*, *A. ruber*, *A. ostianus*, *P. variable*, *P. frequentans*, *P. citrinum* and *Rhizopus* (Codner et al, 1963; Kulik and Holaday, 1967; Scott et al, 1967). It is of interest to find that some strains of *A. niger* produce only aflatoxin B_1 (Kulik and Holaday, 1967).

Strains of *A. niger* are a worldwide distribution in tropical and subtropical areas. In addition to aflatoxin B_1 production, it was found that these fungi have a capability in producing oxalate (Wilson, 1961). Injection of the culture filtrate (7.44 mg/ml) from *A. niger* grew in liquid medium to mice caused tetany, convulsion and death in 5-30 minutes after administration. The LD_{50} of oxalate in mice (20-25 g) was about 3 mg/kg when administered intraperitoneally.

Another group of toxic polypeptide metabolites called malformins was also reported. Malformins are a group of cyclic pentapeptides produced by *A. niger* which cause malformation in bean plants and curvatures in corn roots (Curtis, 1961; Takahashi and Curtis, 1961). Five malformins have been identified. Malfor-

mins A_1 and A_2 were produced in culture filtrate of A. *niger*, A. *ficuum*, A. *awamori* and A. *phoenicis* (Iriuchifima and Curtis, 1969). Malformin A_1 consists of cysteine, valine, leucine and isoleucine in ratio of 2:1:1:1 whereas malformin A_2 consists of cysteine, valine, leucine or isoleucine in ratio of 2:2:1. Malformins B_1 and B_2 were produced only by A. *niger* (Takeuchi et al, 1967). They have a similar amino acid composition except allo-leucine. Malformin B_1 consists of cysteine, valine, leucine and allo-leucine in ratio of 2:1:1:1 whereas malformin B_2 consists of cysteine, valine and leucine in ratio of 2:2:1 (Marumo, 1961). Recently, a new member of malformins called malformin C has been isolated from a A. *niger* van Tieghem, which originated from mold-damaged rice in Thailand (Anderegg et al, 1976). It consisting of cysteine, valine, D-leucine and L-leucine in ratio of 2:2:1:1. According to the toxicity of malformins B_1 and B_2, it was found that they have the same property as malformin A_1 and A_2 (Marumo, 1961). LD_{50} values of malformin C were 0.90 mg/kg in newborn and 0.87 mg/kg in 28-day-old rats treated intraperitoneally. The mechanisms of toxicity are under progressive investigation. Moreover, malformin C showed antibacterial activity against a number of gram-positive and gram-negative bacteria of *Staphylococcus aureous*, *Streptococcus faecalis*, *Proteous mirabilis*, *Sarcina lutea*, *Bacillus subtilis* and *Bacillus megaterium* (Kobbe et al, 1977). In addition, malformin C was highly produced on white wheat and white corn and its production was decreased on glutinous rice.

It has been recognized that investigation of the toxicity of mycotoxins produced by fungi on human foods is very important in general. In Thailand, various market foods and foodstuffs are

highly contaminated with A. *niger* which may produce oxalate and malformins. Accordingly, the high toxicity of malformins may probably create the public health hazard to animal and man. In addition, these strains of A. *niger* may produce a novel mytotoxin. Therefore, it is of interest to screen the acute toxicity of the crude toxins produced by various strains of A. *niger* isolated from market foods and foodstuffs in Bangkok (Glinsukon et al,1975) and to characterize one of the possible new mycotoxin produced from selected strain of A. *niger*.