

## DECONTAMINATING MYCOTOXINS IN AGRICULTURAL PRODUCE FOLLOWING BIOREMEDIATION APPROACH

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### Abstract

*Mycotoxins are secondary metabolites produced by certain species of fungi on grains in the field or after harvest and during transport and storage, etc. These are becoming ubiquitous pollutants in agricultural products, and represent a potential threat to human as well as animal health. Laws enacted to control the presence of mycotoxins in food and feed are increasing. Although some physical and chemical methods of detoxification are reported, bioremediation is going to be method of choice due to its environment friendly nature and low cost involved. Bioremediation is the elimination or biotransformation of mycotoxins into non-toxic metabolites by microbes such as bacteria or fungi. The genes involved in the degradation of mycotoxins by microbial activity have been reported to be cloned, and microbial trials for the treatment of contamination of agricultural products are in progress. This paper briefly describes the toxicity of mycotoxins i.e. deoxynivalenol (DON) and aflatoxin. In addition, the escalation of microbes such as bacteria and fungi, capable of detoxifying these toxins in mixed cultures or pure culture is described. However, the results obtained so far can only be used as a first step in the development of technologies and business practices, as the experiments were performed on a laboratory scale only so far. Finally, future challenges and innovative strategies for decontamination of mycotoxins by microorganisms are elaborated.*

**Keywords:** Bioremediation, Cereal Grains, Mycotoxins, Food Safety, Public Health.

### INTRODUCTION

Agricultural produce are potential host for the contamination of fungi in field as well as during storage, especially in the climatic conditions of Pakistan. Some of these fungi produce secondary metabolites, mycotoxins, which are potential threat to food and feed safety. Mycotoxins may be carcinogenic, mutagenic or may cause interference to hormonal functions in the body of consumer. In advance countries with following industrialized farming systems and sophisticated food processing technologies, mycotoxins may not be a serious threat, however, in developing countries like Pakistan, mycotoxin residues in agricultural produce pose potential risk and may be a cause of chronic illnesses. For example, Ochratoxin may be a cause of renal cancer, deoxynivalenol (DON) can cause human IgA nephropathy, while zeralenon may cause oestrogen in human beings (Creppy et al., 1998; Rotter et al., 1996; Price and Fenwick, 1985). Similarly, presence of these mycotoxins in animal feed may present

serious problems in live stock production, their meat and milk etc. Public awareness about mycotoxins contamination is increasing day by day due to number of reasons. Firstly, world has been global village and people have access to latest information about health and environment issues. Secondly, number of sophisticated techniques for the analysis of mycotoxins have been developed, and general public is conscious about the safety of the food they are going to consume. As, prevention of the contamination of mycotoxins is not practicable, the scientist are putting much more concentration over their decontamination technologies. Though, number of physical and chemical methods have been reported for the detoxification of mycotoxins in agricultural produce, biological methods are of first choice due to their cost effectiveness and environment friendly behavior (Bhatnagar et al., 1991; Park, 1993). Current study deals with latest development for microbial decontamination of mycotoxins in agricultural produce.

## **PREVENTION STRATEGIES AGAINST MYCOTOXIN CONTAMINATION.**

The best pro-active approach to avoid mycotoxins contamination in agricultural produce is the adoption of good agricultural practices (GAPs), which may serve as primary defense line, followed by the good manufacturing practices (GMPs) during handling, transportation and storage etc. The further line of defense may be the practice of Hazard Analysis and Critical Control Point (HACCP) during processing and production systems. However, adoption of these strategies mainly depends upon the local environmental conditions, culture and cropping systems. This is the reason that much attention is paid by the researcher on the solution of problem by developing novel decontaminating technologies, so that the potential health risk associated with the presence of toxin could be minimized. These strategies mainly focus on the:

- Inactivation of mycotoxins or their transformation into non-toxic products
- Destruction of fungal spores and mycelia, so that re-production of toxins could be avoided
- Sustainability of nutritional value of foods after the application of decontamination techniques
- The process should be economical, and easy to apply

## **MICROBIAL DETOXIFICATION OF MYCOTOXINS.**

To meet the above mentioned criteria, researchers have paid much attention on discovery of microbes having potential to biodegrade the mycotoxins. Microorganisms are being screened from different niches capable of transforming the mycotoxins into less toxic compounds (Schatzmayr et al., 2006). This approach called bioremediation is attracting much attention due to its good results and environment friendly, as well as low cost attitude. Some microbes have already been investigated, such as *Flavobacterium aurantiacum* is capable of detoxifying aflatoxins (Ciegler et al., 1996), *Phenylbacterium sp.* capable of degrading ochratoxin (Wegst and Lingens, 1983), and *Gliocladium roseum* capable of detoxifying zearalenone (El-Sharkawy and Abul-Hajj, 1988) has been reported already. We will discuss two major examples in details in the following discussion.

**DETOXIFICATION OF DEOXYNIVALENOL (DON).** DON is a chemically stable mycotoxin, and most commonly produces on

the cereal crops. A large number of strains (1285) were isolated and screened for their DON degrading capability by Volkl et al., (2004). A mix culture was found capable of transforming DON into 3-Keto-DON, and five other unknown metabolites which showed less toxicity than DON. Shima et al. (1997) also reported a microbial strain, *Agrobacterium rhizobium* E3-39, capable of transforming DON into 3-Keto-DON under aerobic conditions. The enzymes responsible for degradation were found in the cell cultures, as well as in cell free filtrate, while absent in cell extract, showing that these were extra-cellular enzymes. Six agricultural soil based strains were reported by Zhou (2008) capable of transforming more than 87% DON from culture media, while two of them were capable of complete removal of DON from culture media. Studies have shown the effectiveness of rumen cultures on the biotransformation of DON. Microbial culture from rumen fluid of dairy cow transformed DON into de-epoxy DON (Yoshizawa et al., 1983; He et al., 1992). Eubacterium strain BBSH 797 was isolated from a rumen fluid, and is most extensively studied DON transforming strain, which also formed a base for a commercial feed additive available in market (Binder et al., 2000b; Schatzmayr et al., 2006). Microflora from chicken intestines has also shown considerable DON degradation activities (He et al., 1992). However, considerable variations have been observed depending upon the breed of chicken, individuals and intestinal regions. Though several authors reported the DON degradation activity of ruminal and chicken intestinal microflora, however Binder et al., (2000a) first time reported a pure bacterial strain, capable of degrading DON. Similarly, Awad et al., (2004, 2006), reported a *Eubacterium sp.* DSM 11798 capable of compensating adverse effects of DON in poultry. However, rumen and intestinal microflora are strictly anaerobic in their functions. So, research on aerobic microbes is still in progress.

## **DETOXIFICATION OF AFLATOXIN**

**(AFB).** Aflatoxins are mainly produced by *Aspergillus flavus* and *Aspergillus parasiticus*. Aflatoxin B1 (AFB1) is the most toxic, mutagenic and carcinogenic, posing most serious threats to animal and human health causing

huge economic losses worldwide. Now a day, much work is being carried out on microbial detoxification of AFB1. Many fungal and yeast species has been reported capable of detoxifying AFB1, such as *Pleurotus ostreatus* (Motomura et al., 2003), *Trametes versicolor* (Zjalic et al., 2006), *Rhizopus sp.*, *Mucor sp.* (Varga et al., 2005), *Saccharomyces cerevisiae* (Shetty and Jespersen, 2006) and *Trichoderma sp.* (Shantha, 1999) etc. Cell extracts of these fungi were capable of detoxifying AFB1, however there were number of limitations found in their practical applications i.e. long incubation time (more than 120 h) and complicated procedures of obtaining their cell extracts etc. Some bacterial species have also been reported as AFB1 decontaminating agents, such as *Lactobacillus sp.* (El-Nezami et al., 1998), *Bifidobacterium* (Peltonen et al., 2001), *Propionibacterium* (El-Nezami et al., 2000) and *Lactococcus* (Pierides et al., 2000) etc. However, it should be noted that decontamination activity was mainly due to binding of toxins with bacterial cells rather than degradation in to non-toxic metabolites. However, bacterial species like *Rhodococcus erythropolis* (Alberts et al., 2006), *Mycobacterium fluoroanthenvorans* (Hormisch et al., 2004) and *Nocardia corynebacterioides* (D'Souza et al., 1998) etc are found involved in biodegradation activity of AFB1.

## CONCLUSIONS

Elimination of mycotoxins from foods and feed still a problem worldwide, and researchers are seeking technologies which could be commercially viable and easy to apply. Microbes can be used as a absorbing agents to decrease the contamination level, as well as biodegrading agents to inactivate the mycotoxins. The main issue to understand in this matter is the mechanism of action of microbes, whether microbes actually degrade the toxin or the toxin is disappeared due to adsorption by microbes? The absorbance just decreases the bio-availability of toxins, while degradation transforms the toxins into non-toxic products. In this regard, first the resistant strains should be screened out, and then study of their mechanism of resistance to mycotoxins may be helpful to understand the biodegradation activity.

Isolation and characterization of enzymes responsible for degradation activity is still under way, and may be a technology of choice, as enzymes offer specific, irreversible, efficient and environment friendly way for the detoxification of mycotoxins. Finally, the development of a technology, which is economically feasible, easy to applicable and friendly to environment is a need of time to decontaminate mycotoxins from agricultural produce, and certainly the microbes are potential candidate for that.

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