World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

AFLATOXINS AND ITS EFFECT ON HUMAN AND ANIMALS: ARTICLE REVIEW

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Article history:

Received: February 28th 2022
Accepted : March 26th 2022
Published: May 6th 2022

Abstract:

Grains, fruits and various foodstuffs are contaminated with fungi and toxins resulting from improper storage before, after and during harvest. The degree of contamination depends on several factors including pH and moisture content, as well as the physical nature of the seeds as well as exposure to insects or microorganisms. Fungi are also characterized by the secretion of enzymes that affect much of the food content. They develop from carbohydrates, proteins and fats accompanied by increased oxidation or staining of food as a result of increased fatty acids that affect the color. The taste and flavor of food . Mycotoxins are a family of biological compounds produced by a group of fungi that have the ability to produce secondary metabolites (SM) when grown in an appropriate environment. The secondary metabolic products of fungi are biologically active compounds. In addition to being non-antigenic toxins, the components that prompt the living body to form antibodies, most of them are toxic."In general, there is agreement that the secondary metabolic products of mycotoxins, as well as the resulting poisoning, should be called mycotoxins. In general, mycotoxins are generally referred to as human and animal food, and the contamination of food or food supplied by fungi, which is called contamination Direct, as food encourages the growth of fungi either during different production stages or during transportation or storage.

Keywords : Mycotoxins, secondary metabolites, fungi , contamination of food

1. INTRODUCTION

The fungus plays an important role in the transformation of the ecosystem as it analyzes the organic matter found in the environment, whether produced from plants or dead animals, transforming complex substances into simple ones that are exploited as nutrients by living organisms and thus provide the basis for the continuation of life and the generation of generations on the surface earth (beretta *et al.,* 2005).

It is also a duplication of work or two-sided. on the one hand, it is of great benefit in the manufacture of different food products and on the other is responsible for the damage of large quantities of food, causing great economic losses.

Some grow and reproduce on food, causing different diseases for the consumer .The beneficial aspect has been exploited by humans. These organisms were isolated and exploited in the manufacture of food products such as cheese, pickles, vitamins, organic acids, enzymes and antibiotics. Yeast was used as an important source of bread, beer and alcoholic beverages.(AL-Sallami *et al.,* 2017).

The economic damage includes the growth in large numbers causing food damage and unwanted odors in the food item, which the consumer may be toxic and harmful to public health .

There are 16 species of fungus that are often pathogenic or food damaged including

Alternaria, Aspergillus, Botrytis, Cephalosporium, Fusarium, Geotrichum, Gleosporium ,

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Helminthosporium, Monilia, Mucor, Penicillium, Rhizopus, Sporotrichum, Thamnidium ,Trichothecium, Brevibacterium Yeast contains 9 genera *Brettanomyces, Debaromyces, Mycoderma, Saccharomyces, Candida, Hansenula ,Rhodotorula, Schizosaccharomyces, Torula.* (Saeed *et al.,* 2016).

The main problem is the ability of a number of fungi to produce toxins (Mycotoxins). It is possible to control the growth of fungi by controlling the conditions for their growth, but it is very difficult to find an ideal way to reduce their production of toxic metabolic compounds produced in developing environments As food becomes unfit for consumption and causes health problems when it is contaminated with fungal toxins, which are known as secondary metabolites of low molecular weight produced by certain types of fungi and have health effects on humans and animals, and lead to food with the effects of the disease its acute effects and chronic effects. (Fink - Gremmels.1999) .

Mycotoxins are a major problem around the world because they affect public health and cause frequent chronic effects, as well as their acute effects when exposed to high doses, especially the liver, kidney and immune system, causing renal failure, liver cancer and immune system weakness (Richard, 2007 ; Ali , 2017).

Mycotoxins are defined as a group of biochemical compounds produced by certain fungi that are capable of producing secondary metabolites when they are grown in an environment suitable for their production. These compounds are bioactive, non-antigenic toxins. Their molecular structure lacks the components that induce the body to form antibodies, most of which are toxic to other organisms. (Ismail, 2014) .

In addition, the secondary metabolic products are separated from some genetically capable fungi to produce toxins when appropriate conditions are available. The Mycotoxins are one of the strongest known toxins that cause serious diseases and at low concentrations of less than 10 ppm. The reason is that they are resistant to heat to a degree that is difficult to destroy when using conventional thermoelectric processes in cooking. fungi can produce more than one poison of the various fungal toxins that cause risks to humans and animals. (Risen *et al.,* 2017).

Also, their effect does not appear quickly but has a cumulative effect and cannot be easily removed from the body and get rid of them, sometimes showing their effect from 10-20 years of eating food and do not seek the immune system in the body to be detected and there is no Therapy drug to reduce the impact and thus pose a big problem. (Risen *et al.,* 2018).

mycotoxins derived from the Greeks Mykes and meaning of the term fungus and Latin Toxicaum , to express secondary metabolic compounds (NRC, 1982) .

Since the beginning of the 1800s, the concept of mycotoxins has been limited to those secondary metabolic compounds produced by fungi in the media that growth on , and no important composition of body the fungus is, This concept includes antibiotics produced by fungi (Cast, 2003). But in 1885 The concept of mycotoxins is of great interest as a result of the case of Ergotism in the treatment of wheat grains contaminated with stone of *Clavicepis purpura*. This condition was accompanied by remarkable symptoms such as general weakness, infertility, low fertility and death for human and animals (Risan and Muhsin2015) .The fungal toxins and the resulting cases remained known and known to the exact scientific level before the year 1960, despite the fact that many unknown cases have caused the cause for many years, leading to many deaths in many parts of the world (Mislivec, 1981). Certain classes of the year and geographical areas are isolated and isolated and therefore have brought little attention to them.

mycotoxins were discovered in 1961 after a disaster in England involving the loss of 100,000 turkeys as a result of their consumption of field pistachios. This case was called Turkey disease due to unknown cause. The disease led to the birth of toxicology. (Mislivec, 1981;Ciegler *et al.,* 1983). An alcoholic extract was obtained from this pistachio which resulted in a case of poisoning of duck birds when treated with it and found that this extract has the effect of brilliance. *A. flavus* was isolated from the pistachios, which proved to be the cause of the disease (Turkey X - disease) and gave the name Aflatoxins for these glittering materials.(EFSA, 2009).

More than 25% of the world's food is polluted with 25-20 micrograms / kg of food. and 4.5 billion people in developing countries are exposed to acute poisoning from contaminated food with high concentrations of mycotoxins (Porter, 1995).

The levels of contamination of mycotoxins are closely related to the environmental conditions in which the crop is grown as well as to the methods used for the transport and storage of such crops (Jonsyn-Ellis, 2000). Growing crops in humid wetlands are more likely to be contaminated with mycotoxins than those on line (Paramare *et al.,* 1997).

aflatoxins was found in a variety of crops, foodstuffs and fodder when it was infected with fungus producing, mainly wheat, barley, maize, soybeans and cottonseeds. These toxins were found in the milk, meat and eggs of animals fed on foods contaminated with aflatoxins and in Iraq (Al-suheily *et al.* 1986) refer in a study of some foodstuffs in the

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Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

local markets confirmed the contamination of wheat flour and rice with high amounts of aflatoxins B_1 while chickpeas and maize were less polluted. *A.flavus* was one of the most isolated fungi (Takahashi *et al.,* 1989).

Many of the seeds of different agricultural crops and their grains are exposed to fungal infections in the field during harvesting, transport and storage operations, especially species belonging to the *Aspergillus*, *Penicillium, Fusarium and Bipolaris* species (Michael and Peder 1982).

Although efforts have been made by researchers around the world or by relevant organizations to reduce the risk of mycotoxins by developing effective plans to reduce food contamination with these toxins, most of these attempts have remained limited and have not been successful solutions to solve this problem This is due to the difficulty of controlling the environmental factors that help the growth of fungi and the production of toxins as well as the intrinsic factors of fungal strains represented by the abundance of genetic changes to them, making control is very difficult . (Kovacs, 2004).

Aflatoxins are a group of mycotoxins, which are toxic metabolites produced by fungi during their growth in various food and cereal crops (Ciegler *et al*., 1981). These metabolic compounds have relatively low molecular weights, where contamination of human and animal feed from field to consumer .

The human exposure to aflatoxins leads to serious diseases according to the dose exposed to it as liver cancer, bone abnormalities, embryonic mutations and fetal deformation as well as its effects on sexual efficiency. In a study in some regions of Asia and Africa, there was a significant correlation between human cancers and the contents of food contaminated with aflatoxins (Carlile *et al.,* 2001; Coker *et al.,* 1984)

Aflatoxins are a group of non-protein compounds that give glowing colors when separated on chromatographic plates and expose the plates to ultraviolet light, giving a bright glow, as shown in the separation of Aflatoxins B_1 ($C_{17}H_{12}O_6$) and B_2 ($C_{17}H_{14}O_6$) and gives two other compounds green emitted with Aflatoxin G_1 and G_2 and another type of aflatoxin known as milk toxins were isolated from cow's milk fed on diets contaminated with fungi produced from aflatoxins. Milk toxins included aflatoxin M_1 and M_2, which were metabolites of both aflatoxins B_1 and B_2 (AL-Dulaimi, 1988) .

Currently, there are 18 types of aflatoxins. B_1, B_2, G_1 and G_2 are the most common and differ in their toxicity depending on their chemical structure and molecular structure. Aflatoxin B_1 is the most toxic and danger toxic (Gold blatt; 1969; Carlson *et al*., 2002).

Aflatoxin is produced by a group of fungi, most important of which are *A.flavus*, which produces Aflatoxin B_1 and B_2 and *A.parasiticus*, which is known for its production of Aflatoxins B_1, B_2 , G_1 and G_2 , and other fungi that produce aflatoxin B_1, but to a lesser extent *A. niger*, *A.wentii* and *A. ruber* and species of *Penicillium* species of *P.citrinum*, known as Aflatoxin B_1, *P.frequentans* and *P.puberulum*, and *Rhizopus spp* (Wood, 1998) .

Aflatoxin is different in its toxicity depending on its chemical composition and molecular structure. Aflatoxin B_1 is the most toxic . The LD50 dose for young ducks at one day is 0.36 mg / kg while B_2 is 1.69 mg / kg (Blatt, 1969).

aflatoxins are excreted by a group of fungus, most notably fungus A .flavus, which produces both aflatoxin B_1 and B_2 (Orum *et al.,* 1999).

Lillehoj *et al.* (1987) note that the fungus mentioned produces aflatoxin B_1 in very large quantities compared to other fungi.

Aflatoxin toxins are described as responsible for serious human and animal diseases such as liver cancer, genital mutilation, abortion, hemorrhage, general weakness and skeletal malformations, as well as their toxic effects in animals, reduced productivity and increased viral and bacterial infections due to the weakening or breakdown of the immune system. (Smith *et al*., 1994)

The aflatoxin is responsible for the emergence of many cases of acute poisoning in humans, with about 500 cases recorded in a number of villages in India and the incidence of fatal liver disease as a result of eating yellow corn contaminated with aflatoxins in the amount of 15.6 to 25 mg / kg, and in Taiwan The consumption of rice contaminated with Aflatoxin B_1 Approximately 0.2 mg / kg to the appearance of several diseases including tumors in the lower limbs with abdominal pain and vomiting (Roberts *et al.,* 1995).

Although fungicides are exposed to several types of contaminants, such as pesticides, antibiotics, and bacterial toxins, fungal toxins are the most severe and dangerous to the variability of the fungal species produced first and the difficulty of detection and control of them as thermoplastic, chemical and physical compounds (Fung and Clark, 2004) , Not only are their effects on health, but they have costly economic effects when they are in agricultural crops. For example, in the United States, the cost of losing cereal crops has been $ 2.5 billion a year as a result of containing more toxins than the limit Which are permitted under US legislation (Wu, 2004) .

In addition, the production of genetically controlled toxins is directly dependent on nutrients and

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

environmental factors (Bullerman, 1981). In addition, the production of toxins is a major factor in the growth of fungi and their production of toxins .

When the fungi grow on the food with the appropriate conditions of temperature, humidity and pH, it begins to grow and then the fungi analyzed the material developing by the enzymes analyzed, converting the complex materials to a simple and when the fungus use these simple substances, either converted to primary products such as cellulose and compounds other carbon-based containers are used by fungi or other microorganisms for the purpose of growing and multiplying, or turning them into secondary metabolic products, some of which are toxic, called Mycotoxins. Fungi produce these compounds when they are less likely to grow While some of the causes of fungus production of these compounds remain unknown, some are a means of defense against fungi that compete with them and are present in the middle of growth or provide protection for fungi from conditions unsuitable for growth such as drought and cold, or used as fungi as food alternatives when depleted The middle ground. (Moss, 2002; Risan *et al.,* 2018).

In addition, heat and humidity play a major role as the *A.flavus*, which affects grain crops in warehouses and is contaminated with toxins, especially during bad storage of heat and humidity suitable for growth, especially thermal range (10-50) m and high humidity, especially at the beginning of storage ,This fungus produces aflatoxin in two days at 21 ° C and 20% moisture (Payane, 1992).

2. THE TOXIC EFFECTS OF AFLATOXINS

Aflatoxins has been known to be responsible for the development of cancers, especially liver cancer, where many infections have occurred in a number of African countries, in addition to their toxic effects on genes, the process of cloning, building DNA and RNA, affecting the building of amino acids and thus inhibiting the formation of proteins (Cocker *et al.* 1984 ; Sahlab *et al.,* 1990).

In view of the seriousness of these compounds on human and animal health, international standards have been developed that do not allow the consumption of food from human hands if it is contaminated with any part of aflatoxins, while allowing some of them to eat parts of it in the animal's animal (Baqir, 1984).

It is estimated that 45 million people in developing countries are exposed to amounts of aflatoxin affecting the human immune system. for example, many cases were recorded in Japan in 1891 after ingestion of fungal-contaminated rice. In 1930 there were symptoms of poisoning of people who had eaten fungal toxin and in 1940 the disease was known as Yellow Rice Toxicosis *Aspergillus* and *Penicillium* as a disease caused by fungal infections and toxins (Nakhilan ,2011) , and in Russia died about 40-50% Added to what happened in July 2004 in Kenya from cases of poisoning Which killed 125 people Singled out 317 (Walliams 2004 , Lewis *et al* 2005).

Studies have shown that human and animal exposure to aflatoxin toxicity leads to serious diseases, such as liver cancer, bone abnormalities, genetic mutations, fetal malformations, as well as their effects on sexual function and immune system (AL-Heti, 1992). Aflatoxins are considered to be responsible for the development of cancer ,This effect was discovered during epidemiological studies conducted in some regions of Asia and Africa. The significant association between human cancers and food contents of aflatoxins was observed (Cocker *et al.,* 1984).

The World Health Organization's Global Agency for Research on Cancer reported that aflatoxins are among the known causes of human cancer, adding that 25% of the world's food and feed is at risk of contamination with fungal toxins (Watkins and Curtis, 2003, Park *et al*., 2009; Melvin, 2012) .

There is strong evidence to confirm that the carcinogenesis of these compounds is through genetic poisoning, which involves metabolic activation of the composition of the epoxide and a modification of the gene responsible for inhibition of tumors TP53 (Tumor-Suppressor gene) by replacing G with T of the nitrogen bases in this gene .In other types of aflatoxins, an interaction of aflatoxins occurs with the DNA molecule at the seventh nitrogen atom (N-7) . (AL-Hajar , 2018) .

Aflatoxins affects white rat embryos during pregnancy When exposed to Aflatoxins B_1, pregnant women were exposed to cancerous tumors in newborns that had contaminated milk from mothers who had fed a contaminated diet. It also caused many congenital malformations in the fetuses as well as high mortality In the case of infertility, it can cause many disorders of sexual characteristics and immunity inhibitors. The administration of poultry doses of aflatoxins ranging from 0.25-0.5 mg / kg in feed causes a decrease in the immunity of birds against bacterial and viral infections (Ibrahim and AL-Jabouri, 1998).

A study conducted by Hamoudi and Al-Dori (2001) found when give food contamination with Aflatoxins B_1 for chicken that the administration of high levels of internal organ weights (spleen, heart, stomach, and gout) was significantly higher in the weight of the liver because it was the fastest affected by mycotoxins.

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
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Abu Shaba (2003) noted that *A.flavus* and *A. niger* had a satisfactory effect on the tissues of the white mice, which resulted in severe changes in the chromatin material and an abnormal increase in the size of the nucleus accompanied by congestion of the bronchial tissue of the liver and its effects on the tissues of the kidney, other study of the toxic effects of *Aspergillus parasiticus* and *Bipolaris micropus* showed a reduction in the weight of animals treated with fungi where 10.2% and 7.2% respectively had a effect on blood parameters. The results indicated that the number of kidneys W.B.C was raised in the blood of animals treated by *A.parasitcus*, which reached 6600 cells / ml while it reached 10500 cells / ml in the blood of animals treated with fungus *B.micropus*. and the same time, ESR values did not change in all treatments, Elevated Hb values were 15-18% for animals treated with A. *parasiticus* and *B.micropus*. In addition, they affected the liver, where liver tissue degeneration occurred with the death of Necrosis cells. The other kidneys were also affected by the toxins produced, (mohsen *et al* , 2009).

On the other hands , AL- Musawi (2010) , AL-Khalaf (2011) reported that aflatoxins had a satisfactory effect on the tissues of the animals, causing severe tissue changes, such as congestion, bleeding, as cites in the arteries and veins of the liver (hypertrophy), as well as inflammation of the tubules, necrosis of the intestine and bloody hemorrhage. In 2006, mohsen studies Laboratory animals were treated with wheat grains contaminated with aflatoxin. They were low in body weight, especially *A. niger* and *A.flavus* . Low body weight in general. There was a decrease in the number of primary and secondary ovarian follicles graffian follicles as well as ovarian diameter which reached to 26.0 and 15.6mm , The ovaries train, in addition to an increase in uterine weight and thickness, and some of the animals got infertility, where there was no pregnancy in females for a period of time up to three months when mating with males together.

AL-Jubouri (2012) showed that the effects of aflatoxin B_1 and B_2 on the reduction of levels of FSH in the blood of treated animals were affected separately. The rate of this hormone in the blood of animals treated with B_1 was only 2.7 ng / ml with a significant decrease of p 0.05 compared to the control treatment of 4.1 ng / ml when the level of this hormone in the blood of animals treated with poison B2 only to 3.9 ng / ml.

3. BIOINFORMATICS AND METHODS OF CHEMICAL TRANSFORMATION OF AFLATOXINS

The aflatoxins are exposed to different metabolic processes and the efficiency of the organisms in the elimination of toxin is differentiated by metabolic processes. The liver microcosms are able to convert aflatoxin B_1 and G_1 toxic to non-toxic B_2a and G_2a compounds. The appearance of M_1 in human urine and many animals is found to be related to the conversion efficiency of toxins B_1 and B_2 to M_1 and M_2 are less toxic, and M_1 isolation from cow's milk is also a pathogenesis of B_1 (Roberts, 1990).

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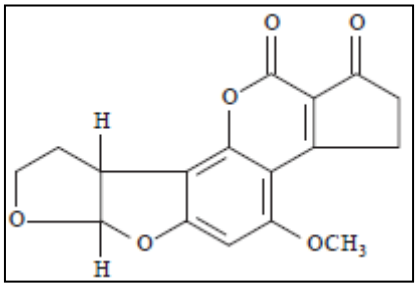
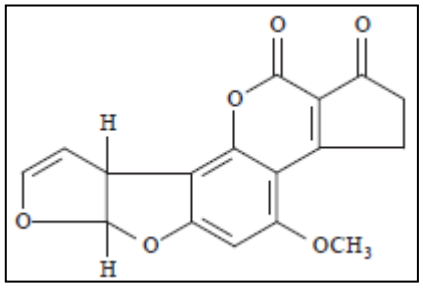
Volume-10, MAY 2022
ISSN: 2749-3644

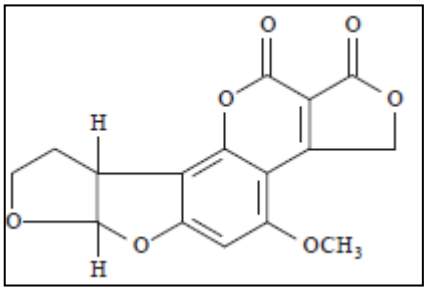


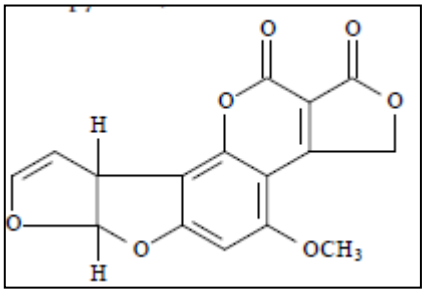
Aflatoxin B_2



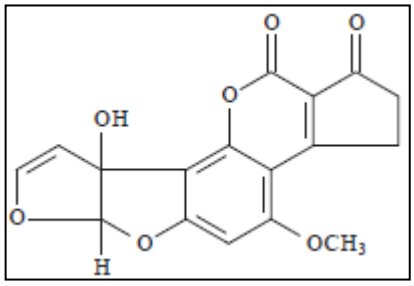
Aflatoxin B_1, $C_{17}H_{12}O_6$

$C_{17}H_{14}O_6$





Aflatoxin G_1, $C_{17}H_{12}O_7$ Aflatoxin G_2 $C_{17}H_{14}O_7$



Aflatoxin M_1 $C_{17}H_{12}O_7$

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

Figure (1) Structure of type aflatoxin

The production of secondary metabolites of toxins depends on the type and genetic composition of the fungi and the nature of the chemical and physiological environment in which the fungus occurs. Although some strains have the genetic ability to produce certain types of fungal toxins, the amount of toxin produced is affected by the amount of nutrients available in the developing medium In addition, the presence of amino acids, such as gluotamic acid and Aspartic acid, improves the synthetic processes of the production of poison. The presence of zinc is essential for the formation of these compounds .In addition to dietary requirements, physiologic factors such as temperature and water efficiency are instrumental in the process of producing these compounds. (Risen *et al* ,. 2017)

aflatoxins are pale blue crystalline compounds that are pale yellow, low in water and hydrocarbons, dissolve in methanol, acetone, chloroform, dehydrate and non-polar solvents, are stable at temperatures below 100 m and in the absence of light, relatively unstable in light and air When exposed to oxidizing agents or ultraviolet light or solutions that have a pH of less than 3 or more than 7.

Fused between 273 m for G_1 to 299 m for M_1. However, they do not break down in normal cooking conditions but are completely shattered by autoclave with ammonia or when treated with a deficient substance. (AL-Hajar, 2018).

It is characterized by its brilliance under ultraviolet (UV) and is therefore classified as B and G depending on its brilliance under UV and along the wavelength (365) nanometer, where B takes from blue and means blue brilliance, G is taken from Green , which means green brilliance and type M because it is present in Milk.

Figures 1 and 2 written beside the letters refer to the separation sites of these compounds on the Thin Layer Chromatography (TLC) separation plates. .(Meerdink,2004).

Fluorescent glare is strongly characterized by exposure to long wavelengths of ultraviolet radiation, which makes it possible to detect the presence of toxic compounds at very low levels (0.5 ng or less for each of the detection spots on the chromatographic plates.

The presence of the Lactone ring in the aflatoxin molecule makes it more susceptible to hydrolysis in basal solutions. This is an important feature in the process of the manufacture of food with rules that reduces the level of contamination of aflatoxin in the final product. Table (1)

Table (1) Physical and chemical properties of aflatoxins (Eman, 1999)

Aflatoxin	**Molecular formula**	**Molecular weight**	**Emission glow when exposed to UV**	Coefficient of RF
B_1	$C_{17}H_{12}O_6$	312	Blue	0.56
B_2	$C_{17}H_{14}O_6$	314	Blue	0.53
G_1	$C_{17}H_{12}O_7$	328	Green	0.48
G_2	$C_{17}H_{14}O_7$	330	Green	0.46
M_1	$C_{17}H_{12}O_7$	328	Blue	-
M_2	$C_{17}H_{14}O_7$	330	Blue	-

Described the chemical composition of these types of aflatoxins according to NTP in America: known as a human carcinogen and IARC in France: within group (1) a confirmed carcinogen of human .(AL-Hajar ,2018).

There are many differences in the biological transformations of aflatoxins within the body of an organism between different species in general and sometimes in one species.

There are four metabolic pathways of aflatoxin B1 within the body:

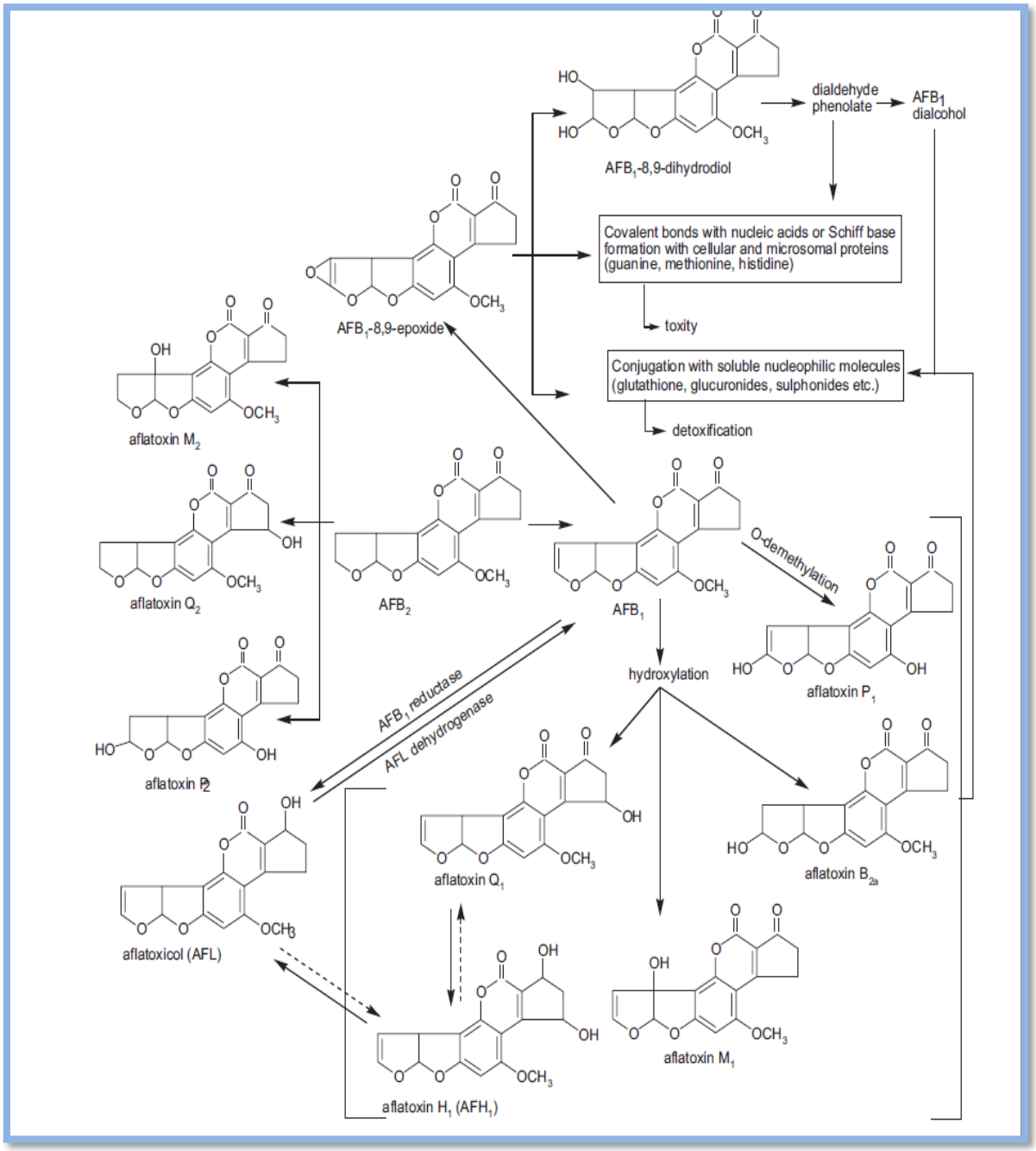
1. O-de alkaylation into aflatoxin P1 (AFP_1).
2. Ketoreduction into aflatoxicol (AFL)
3. epoxidation into AFB_1-8,9 epoxide
4. hydroxylation into AFM_1 or AFP_1, AFQ_1 and AFB_2a.

The most common type of compounds 8,9epoxide is toxicity, mutations and cancer and AFM_1 is highly toxic, while AFP_1 and AFB_2a are relatively nontoxic compounds, as shown in figure (2) described by YianniKouris and Jouany (2002).

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

The metabolic differences of aflatoxins within the body of the organism are AFB_1 to AFL, which is the primary metabolic product of duck liver, whereas AFB_1 is AFQ_1 and AFB_2a in ducks, rats, mice and monkeys except humans (Robuck and Wogem, 1977).
Chung *etal*,(1998) found that AFB_2a and AFM_1 are also metabolites in chickens.
Figure (2) Metabolism of aflatoxin B_1 and B_2 within the body of the organism (Yainnikouris and Jouany, 2002).

Aflatoxin is subjected to many metabolic processes. Studies show that aflatoxin B_1 is converted into several secondary metabolites, one of which is M_1, a highly toxic compound. Despite the toxicity of this secondary compound, the body has the potential to neutralize toxic action by binding to the yellow gland acid In the liver before being put into the yellow bag or with the urine, as well as newly discovered that there are two secondary metabolites P_1 and Q_1 are undergoing the same transformations to neutralize the toxic effect. (Yainnikouris and Jouany, 2002).

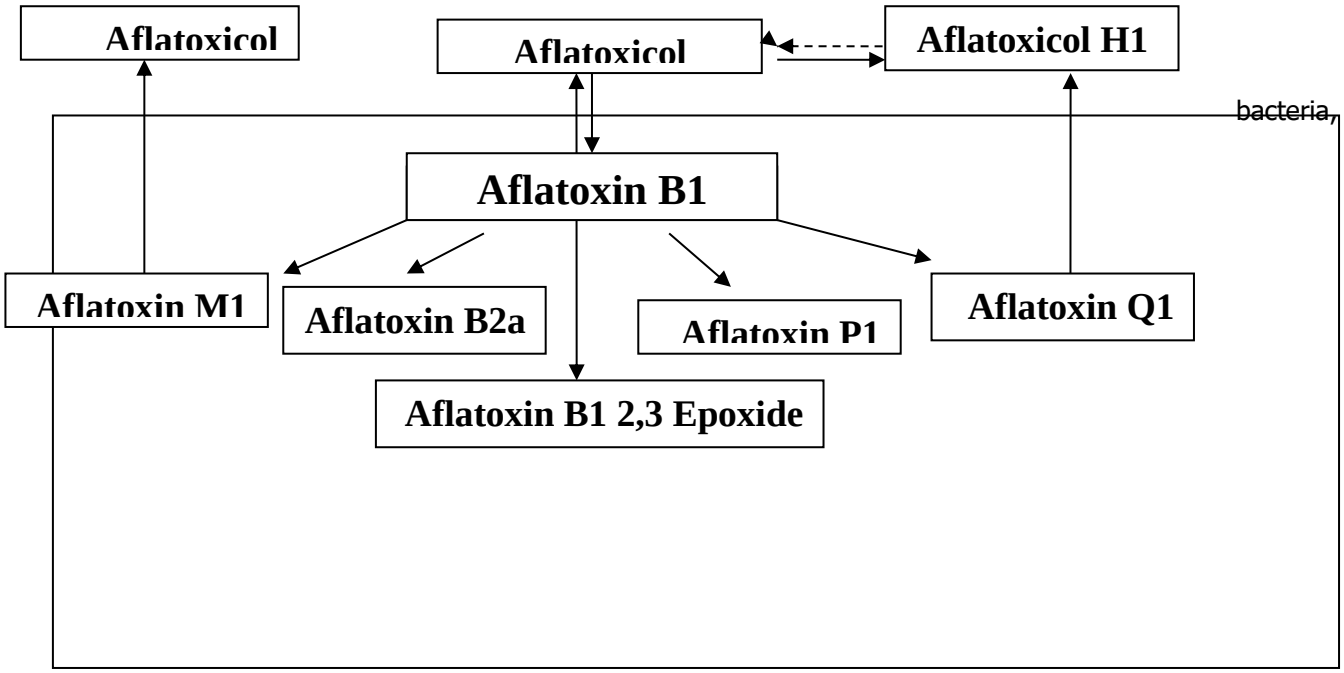


World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

Figure (2) Metabolism of aflatoxin B_1 and B_2 within the body of the organism (Yainnikouris and Jouany, 2002).

In the liver, Aflatoxin B_1 is converted to aflatoxicol and then to aflatoxicol H1. Aflatoxin B_1 can also be converted into the secondary aflatoxin Q_1, which in turn can be transformed into aflatoxin H_1. These changes are made by helping enzymes found in the liver. Liver cells of some species of birds and rodents have been found to be very effective in converting Aflatoxin B_1 and G_1 into secondary metabolic compounds called B_2a and G_2a. These compounds have high protein binding ability and cause many of the severe adverse effects associated with the toxicity of these compounds. (Ibrahim andAL- Jabouri, 1998). The following figure illustrates the biochemical transitions of adenosine in the liver.

Figure (3) illustrates the biochemical transitions of adenosine in the in the liver.



4.ROLE OF MICROORGANISMS IN INHIBITING FUNGI OR REDUCING THE EFFECT OF AFLATOXIN PRODUCED

The interaction between fungi produced from toxins and microorganisms is common in nature as it affects the inhibition of the growth of fungi and its production of toxins. The use of microorganisms has been common in the last two decades as scientific development has contributed to this and its entry and competition for chemical control. The endemic bacteria were used in soils for the production of antibiotics Against the fungus and reduce its toxic limit. (Al-Ashour, 2005).

The efficiency of many types of microorganisms has been tested in inhibiting a range of fungal .(Ciegler *et al.*, 1964) tested the efficacy of several microorganisms, including a number of fungi and and studied their effect in inhibiting the growth of fungi producing aflatoxins, especially aflatoxin B_1 and G_1, in many crops .

Doyle *et al.* ,(1982) refer to a group of organisms including bacteria *Corynebacterium rubrum* and fungi *Rhizopus oryzae* and *Trichoderma viride* have demonstrated their ability to inhibit the growth of aflatoxins producing fungi as well as their ability to convert aflatoxin B_1 to non-toxic aflatoxin B_0 .

The use of non-producing strains of *A.parasiticus* in cotton fields has been significant in causing significant inhibition of toxic strains of *A.flavus* and *A.parasiticus* producing aflatoxins (Dorner *et al.,* 1992) .

Cotty (1994) used strains of *A.flavus* non-productive of aflatoxin toxin to resist and reduce aflatoxins produced by toxic fungal strains when added to the cotton field crop. The addition resulted in a

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

significant reduction in isolated isolates, especially B_1 compared to untreated fields for these breeds.

Lauzon *et al.*, (1996) reported that the treatment of *Cladosporium fulvum* for rice extract in both laboratory and field reduced the incidence of *A.parasiticus* and prevented the production of aflatoxins. The reason was attributed to the ability of *C.fulvum* mushrooms to produce effective substances in *A.parasiticus* and germination, thus inhibiting the production of aflatoxins

A number of tests have also been conducted on the efficiency of microorganisms in inhibiting the microbial growth of contaminated foods and destroying their toxic secondary metabolic products. These microorganisms have the ability to compete with fungi on the base material or to release certain antibodies (Doyle *et al.*, 1982).

The choice of the microbial object of resistance is an important factor before it can be used in the field. It must be tested for its application. This depends on its production of the important antibodies , enzymes and Lysozymes.

Bacillus and *P. fluorescens* have the ability of Antagonism, One of these neighborhoods is to destroy the other organism or limit its growth to maintain its vitality and ability.

for example, bacteria producing Lactic acid LAB and yeast *saccharomyces cerevisiae*, which possesses the yeast ability to produce poisons have the ability to reduce the risk of pollution and the production of fungal toxins in food .LAB is added to food to enhance its quality and increase the value of food (Mahmoud , 2011).

On the other hands , *Bacillus* was found to be one of the most important species in the field of vital resistance to possessing many of its species to the specifications of the most efficient biological factor because it possesses distinct mechanisms that have made it a superior element in the field of bio-resistance and the manufacture of fertilizers and pesticides. This is indicated by many studies Which indicated its success in controlling a range of important diseases affecting strategic crops. For example, the *Bacillus subtilis* GB_{01} strain was used as a commercial powder in the United States under the brand name Kodiak to resist seedling disease in wheat and cotton (Safiyazov *et al.,* 1995). Several tests were carried out on *Bacillus* bacteria, such as *B.cereus, B.pumilis* and others, where research results demonstrated the efficacy of these species in curbing the activity and effect of many plant pathogens and on different crops (Yuming *et al .,* 2003) .

In Iraq, the first study of its on *Bacillus* was carried out in biochemistry applications, which tested the efficacy of *B.cereus* bacteria in controlling wheat and buck worm disease (AL-jumaili *et al.*, 2007, Ali ,2017) .

A number of scientific research has been carried out on the Bacillus species, especially after the species *Bcereus, B.subtilis and B.pumilis* have demonstrated a high ability to inhibit the growth and activity of many pathogens and reduce their economic damage (Dal-soo *et al.,* 1997 ; Yuming *et al.,* 2003) .

Garica and Angles (1994) found that *B.megaterium* bacteria are highly efficient in inhibiting the growth of *A.flavus* and the formation of aflatoxins .The use of the antibiotic Iturin A, a fatty protein produced by *B.subtilis* with a concentration of 50 ppm, inhibited the growth of A.parasiticus and inhibited the production of aflatoxins (Ono and Kimura, 1991) .

In a study conducted by Al-Ashour (2005) and 2009 , which included the production of a biological product of the *B.cereus* vaccine and its efficacy against a number of fungi, including *A.flavus* and *A. niger*, the test results showed its high ability to inhibit the growth of fungi completely 100% on the agricultural medium.

A number of studies have been conducted to test its ability against a variety of fungal pathogens, especially those that cause rotting and seedling diseases (Abdul-Jalil, 2004 , Gali, 2004). Its ability to protect maize grain from *A. flavus* and *A. niger* and the contamination of grains with aflatoxins. This study has demonstrated the ability of the biocide to inhibit the growth of the plant in the cultural medium by 100% when used at a concentration of 1 g / L. The pesticide also provided a high protection for yellow corn grains from infection for a period of time of up to six months, Percentage of infection in cereals for both fungus 3% and 93% in control-treated grains in non-biocidal maize .(Abu Shaba, 2003) .

In the study (Risan and Mohsen, 2015), using the metabolic products of *B. licheniformis* were used to reduce the production of A. flavus aflatoxin B_1.

other study was carried out to evaluate the efficacy of thurine w.p ,
a product of *B.thuringiensis* bacteria, which is loaded with calcium carbonate on the vitality of stored *A.flavus* wheat grains. Several fungi associated with wheat grains were isolated, including A.*flavus, A. niger and A. terreus, Penicillium spp, Rhizopus spp,* and *A.flavus* were the most affected, and their toxicity was 100% compared with the rest of the fungus (Mohsen *et al.*, 2011)

P.fluorescens has the ability to produce a number of antibiotics that inhibit the growth of a number of pathogens called antifungal compuonds such as HCN and Pyrrolinitrin. Abu Shaba (2003) tested these bacteria and obtained the ability of these bacteria to

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

provide protection for yellow corn *A.flavus* and *A. niger* infection in the store as well as its ability to provide high protection for rice grains from a variety of store fungi (Kuhdier, 2005).

Both Al-Jumaili and Abu Shaba (2005) and Mohsen (2006) reported that *Pseudomonas fluorescens* have a high ability to protect the seeds of maize and wheat from *A. flavus* and *A. niger.*

P.flourescens pf-5 was used to inhibit the growth of *A.flavus*, as well as the effect of *A.flavus* on the percentage of infection corn after being fertilized with the extract for 15 minutes, reducing it to 3.3% compared to 100%. The bacterial-treated seeds were not contaminated with aflatoxin B_1 during the six-month storage period (Kassem , 1998).

P.flourescens gave good protection to potato tubers from *Fusarium solani* for three months under natural storage conditions (AL-Khalkhali, 2005).

On the other hand, Ibrahim and AL-Jubouri (1998) noted that the use of *Flavobacterium aurantiacum* bacteria, where this organism in aqueous solution can represent aflatoxin B_1, G_1 and M_1, but has not yet been chosen, the feasibility of such methods at a broad commercial level.

AL-Duhaidahawi (2014) studies the possibility of producing specialized enzymes in the analysis of mycotoxin for use in broad practical application.

5.FACTORS AFFECTING FUNGI AND THEIR SUSCEPTIBILITY TO THE PRODUCTION OF MYCOTOXIN

Environmental factors play an important role in the growth of fungi and their ability to produce mycotoxin include humidity, temperature, pH and others factors:

5.1 Humidity

Moisturizing content of food is an important factor that determines the growth of fungus and the production of mycotoxin. The moisture content requirement varies from one fungus to another, even within the species. Some fungi need moisture content ranging from (13-18) % and (18-20)% others (22-26) % .(Asker, 2004) .

Moisture is one of the characteristics related to the weight of dry matter. For example, *A.flavus* grows in humidity (80-85) % and produces its poison even in the case of low humidity. The process of storage of crops in humid conditions followed by hot conditions and drying period of the most important factors that help fungi to produce (Malmauret *et al.,*2002) .

Many fungi can grow on food stored under relative humidity of up to 90 %. Many fungi belonging to the Mucoraceae family can grow on food stored under relative humidity ranging from (90- 100)% and other species grow at lower levels and some species of *Penicillium* spp grow under humidity levels up to 75% and some species of *Aspergillus* spp grow under levels of moisture up to 65% . (Robert and Charles, 1960) .

5.2 Temperature

Most of the Mesophilic fungi have an optimal temperature of (25-30) ° C, but some grow(35-37) ° C, such as *Aspergillus* spp . (Marybeth ,2000) .

A.flavus grows at a temperature range of (10 - 43) °C. The optimal temperature of growth is limited to (33-32) °C. The production of aflatoxin is at a temperature of (12- 40) °C and optimal for its production (35-25) °C. Aflatoxins at temperature change At low temperature (18-15) ° C, Aflatoxin B and G are produced in approximately equal amounts, while Aflatoxin B production at temperate temperatures (25-28) ° C is predominant at the expense of Afaroxin G.(Faraj ,1990)

On the other hand , that the reduction in Aflatoxin G_1 production is due to increased catabolism at high temperatures (WHO, 1979) .

Mahrous (2006) refer that aflatoxins do not produce at temperatures below 20°C and above 35°C, that the best temperature for the production of aflatoxin type B_1 was 24°C and for the production of aflatoxin of type G_1 at 30°C, while one researcher found that The most suitable temperature for the production of these two types of aflatoxin is 24 and 32°C, respectively.

also found that the time for incubation of the fungus had an effect on the percentage of mycotoxin produced, obtained the highest amount of poison after 5-12 days of growth of the fungi and then followed by a decline again in the production of toxins, and found that the accumulation of aflatoxin in the corn up After 4 days of incubation, followed by a reduction of up to 80% of the maximum quantity produced on the eighth day.(Al –Saedy ,2015).

5.3Water activity

Water activity is a measure of free water that is not associated with the food necessary for the growth of fungi and studies indicate that the water efficiency needed by fungi is much less than that needed by bacteria (.Northolt *et. al.,* 1976)

It is known that when water activity is low, water will be associated with salts, sugars, proteins and other dissolved substances. for this reason, the growth of fungi does not occur when water is not readily available. However, the growth of fungus can occur on values of water activity less than (0.85) that, which is observed for the growth of *A. flavus* where it was found that the least activity in which the water is growing fungus is (0.78). However, the optimal

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

water activity for the production of aflatoxins by *A. flavus* is at (0.95). (Davis and Diener, 1970 ; Northolt *et. al.,* 1976).

The production of aflatoxin occurs in varying degrees of temperature and water activity. Aflatoxin B_1 is formed in conditions where temperatures and water activity are close to the minimum limits for the growth of the fungus (Ibrahim and AL- Jabouri, 1998).

5.4 Oxygen

Oxygen is necessary for the growth of fungi, but some prefer anaerobic growth for the production of ethanol and organic acid, as well as affect the production of mycotoxin, but is limited to the production of aflatoxin significantly if the level of oxygen less than 1% in the center where the fungi producing Aflatoxin. (AL-Duhaidahawi , 2014).

Fungi are highly oxygenated organisms required for vegetative growth, germination, and germ formation. The fungi vary in their ability to tolerate high concentrations of carbon dioxide, and most fungi can only grow with at least 1-2% oxygen .(Asker, 2004) .

In general, reducing oxygen concentration to the production of aflatoxins, and the deficiency is evident when the concentration of oxygen is reduced to 1% with the increase of carbon dioxide to 80%. One researcher found that the production of aflatoxin from the *A. falvus* has decreased significantly with a decrease The concentration of oxygen from 5% to 1% and increased concentration of carbon dioxide from 0.03 to 100%. also found that low oxygen concentration to less than 20% or increased concentration to 90% or higher inhibit the formation of aflatoxin and nitrogen gas to inhibit the formation of aflatoxin .(Amedo, 2007) .

5.5 PH

PH fungi grow naturally in acidic pH center (3.5-4.5) , PH is affected by the production of mycotoxin . If the PH to 4 or less, the production of aflatoxin increases to the highest concentration and reduce the number of spores formed by 50% or less in the *A.flavus* .(Cooty, 1988) and that the production of toxin decreases in both the fungal A.*parasiticus* and *A. nidulans* as the pH of the medium .(Keller *et .al.,* 1997) .

In addition, the higher the pH, the greater the effect of the enzymes of the organic matter, which play a role in the production of toxins (Isabel *et al.,* 2000) while the production of mycotoxin decreases as PH, Keller et al (1997) The aflatoxin excreted by *A.flavus* produces its highest concentration at PH = 4 or less. (AL-Duhaidahawi , 2014).

5.6 Fungus strain

The diagnosis of the type and strain of fungus contaminated to food can be valuable in the case of epidemiological cases of fungal poisoning, and the production of mycotoxin is a process controlled by genetically, and the mycotoxin in the composition and toxicity and type, according to the strain of fungi produced . (Asker, 2004).

The type of fungus is a key determinant of the nature and type of the toxin *A. flavus* produces two types of aflatoxins B_1 , B_2, while A. *parasiticus* produces aflatoxins B_1, B_2 , G_1, G_2 . (Hesseltine *et al.,* 1985). As well as , The strain of fungus and the genotype of the strain is responsible for identifying the innate toxins produced (Moss and Smith, 1985).

Many species that produce aflatoxins, such as A*. flavus, A. parasiticus, A. oryza, A. falvus* and other types of aflatoxins, such as *A. rubber, A. niger, A. wentii, Penicillium puberulum* ,Secondary representation .About 30% of aflatoxin produced from A*. flavus strains, P. puberulum* is one of the most contaminants for agricultural crops and food grains (rice, maize, barley, peanuts, walnuts) and some food products such as bread and dairy products, some fermented products and Stored under appropriate conditions of humidity and heat . (Mahrous ,2006).

6.FACTORS THAT HELP PROTECT FOOD FROM FUNGAL INFECTIONS AND AFLATOXINS

A number of efforts have been made to reduce or prevent fungus and reduce its susceptibility to the production of toxins if a number of methods are used which must affect the organism's physiology and not leave any harmful compounds.

6.1 Physical factors :

Mechanical methods are one of the methods used to eliminate fungus by isolating infected seeds from the apparently uninfected, but they need effort and cost. So, look for other ways (Rustom, 1997) .

Physical factors were used in the treatment of various foods, especially grains and cereals of basic crops such as wheat, maize, rice, barley and others, with heat, radiation and microwave.

Heat is an important factor in this field, as it reduces the moisture content of grains and grains at levels that make them safe from fungal infections in the store (Cocker *et al.,* 1984). Most store fungus needs moisture content ranging from (14 – 19) % depending on grain type and the common occurrence of the moisture content of maize grains is 13% for *A.flavus*, 17% for rice, 12% for cotton and 13.5% for barley (Chang and Markakis, 1981 ; Davis *et al* ,. 1984)

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

Tomas and Athapol (2002) indicated that the yield should be dried so that the moisture content is (12-14) % and should be reduced to 18% for some wheat, barley and maize crops .

Drying is usually done naturally by exposure to sunlight and using air currents that allow moisture to be reduced without affecting the vitality of grains. The drying process is quick to avoid exposure to fungal infections (Cocker *et al.,* 1984).

In the field of mycotoxin, a single study was conducted in which micronutrients were used to break down aflatoxin B_1 in field pistachios. The amount of aflatoxin B_1 decreased from 393 µg / kg in comparison (non-microwave) to 0.6 µg / kg when exposed to polluted grains Minute for five minutes (AL-Jumaili, 1996, Maitree, 2000).

Mohsen and Risain 2009) conducted a study to determine the effect of x-rays, UV rays and gamma rays on the growth of *A.flavus* and *A. niger* in wheat grains .The results showed a difference in the sensitivity of fungus to radiation, where *A.flavus* mushroom was more sensitive to *A. niger* .Moreover, these rays had a clear effect on the ability of fungi to produce aflatoxins, which lost their ability to produce these toxins in comparison with the control treatment that had the ability to produce toxins.

Recently, the laser has been in use as it has been used to sterilize milk to inhibit or reduce bacteria, fungi and yeasts, and to limit their ability to produce toxins without physical and chemical changes in food (AL musawi *et al*., 2018) . As well as the use of NANO technology to eliminate fungal toxins such as silver nano and nano composites in packaging (Ibrahim , 2012).

In addition, storage under low temperatures is appropriate to control the growth of the molds and these methods of commercial storage, but it is not useful because it is likely to get higher humidity and increase the opportunity for growth of the molds.

It is therefore possible to control the prevention of growth of the molds and therefore the production of toxin during the storage period of the grain is to change the environment around the seed itself and control the humidity and temperature in the store, and the disposal of insect injuries as it can damage the outer casing of the grains moving fungal bites from place to place inside Place of storage, so these factors must be taken into account to avoid fungal infections.(Mohsen and Hussein ,2011) .

6.2 Chemical methods

When physical methods are useless to control the growth and toxicity of fungi, it is necessary to direct to chemical means to achieve the said goal.

Chemical methods are one of the most effective means of destroying toxins. FAO has established basic conditions for the use of chemicals in this field. It must be highly effective, do not leave toxic residues, prevent pollution of the environment and preserve the food in terms of structure and taste. The final leaves carcinogenic toxins to the final product, so FAO warned against using them commercially (Bhanuprakash *et al* ,.2011) .

Several chemicals have been used to treat fungus in the store, such as ammonia, as it works to reduce aflatoxin by 99% when performed under appropriate conditions. This technique is safe and potentially poisonous because it can open the lactone ring in the AFB_1. The addition of urea (7 and 10%) to the fodder containing aflatoxin B_1 led to the smashing of the poison after two weeks of treatment (Majeed 1997).

On the other hand ammonia was mixed with ammonium hydroxide with certain conditions of moisture and heat, which was placed on the feed led to the animals refrain from eating feed for the possibility of toxicity and composition of compounds containing aflatoxin B_1 . (Diedhiou *et al.,* 2012).

Ibrahim , (2012) ; AL-Hadad *et al.,*(2016) refer to the use of ozone O3, a powerful oxidizing agent, is highly effective in destroying toxins if it is effective in breaking down aflatoxin B_1 but is ineffective on aflatoxin B_2 .

The gas has been used to inhibit the growth of fungi and reduce their toxicity. This gas has the potential to inhibit the growth of *A.flavus* and *A. niger* when treated at 100 ppm. It can inhibit the growth of fungus filament of *F.oxysporum* at a concentration of 200 ppm. In addition, Caused by *P. italicum* when treated with gas for 7 days (Mahmoud ,2007).

Abdul Azeez , (2015) in his study show the use of ethylene gas at concentrations of 100 and 125 ppm is highly effective in inhibiting the growth of *A.flavus* and *A.parasticus* isolates isolated from maize.

6.3 Adsorption aflatoxins from body

In recently many methods have been proposed as a solution to the risk of aflatoxins based on the deactivation of the aflatoxins directly in the animal's digestive tract where chemicals that have the ability to absorb ate the aflatoxins are added and made unavailable. Several studies have demonstrated in field conditions the ability of the compound sodium calcium aluminosilicates (HSCAS) to neutralize aflatoxins effectively and or may fail to neutralize toxins in a limited way with other toxins such as Trichothecenes, zearalenone, ochratoxin A, fumonisin (Ibrahim 2012) .

6.4 Biological methods for destroying fungal toxins

World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-10, MAY 2022
ISSN: 2749-3644

Despite the rapid and effective results that can be obtained as a result of the use of chemical pesticides in the control of fungal diseases, the expansion of their use has led to the disruption of natural balance resulting in the emergence of diseases not previously known and the remnants of many of them are toxic to humans and animals, The biological balance between microorganisms and the activation of a group of organisms (plants, microbes) in general or in particular against one or more pathogens, known as biological control. Most researchers believe that fungicides are no less effective than pesticides , a plant extracts was found that the bacterium *Bacillus subtilis* with the effect of high antibacterial against the most important pathogenic fungi such as *Fusarium*, *A.flavus*, *Penicillium*, *Rhizioctonia* and *Pythium* (Stover and Driks , 1999; Young *et al.,* 2003 .)

While plant extracts were used to control some pathogens, *Zingiber officinale* was used to control many plant pathogens. Many attempts have been made to investigate active substances in support of environmental efforts as alternatives to chemical pesticides with environmentally desirable properties such as rapid degradation, low toxicity and high specialization (Schutte *et al.,* 2003) and this water extracts using against 18 of fungi type *Penicillium* and *Curvularia* and others (vechet *et al .,* 2009 ; Mohsen 2010) .

In a study conducted by Al-Khalaf (2011), where the ability of honey to reduce the toxic effects of aflatoxins B_1 and B_2 in the biological systems of male white rats proved to be high. The concentration of hemoglobin increased. Honey also played a major role in maintaining the levels of phylogenetic and other biochemical parameters. Normalizing the level of the induction hormones for LH and FSH in serum lipoproteins B_1 and B_2. This substance has a role in protecting liver, spleen and intestinal organs from the toxic effects of aflatoxins .

Another study of the effect of coffee and its content of caffeine in the growth of *A.flavus*, *P. pglabrun* and the production of aflatoxin G_1, and patulin and found that the highest concentration of the use of 1 g / liter of caffeine absolute reduced the growth of fungi, and prevent the production of aflatoxin G_1 and 100% The saffron proved to be an effective in total inhibition of the production of fungal toxins produced by some strains of *Aspergillus* and *Penicillium*, and the *Cinnamon* and Saffron material had a 100% total inhibition of fungal growth and the production of fungal toxins.(Al_Qarawi *et al ,* 2008) .

7. CONCLUSION

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