



Research Article

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Mycotoxin Contamination of Stored Maize in Kenya and the Associated Fungi

Jonah K Birgen¹, Richard C Cheruiyot¹ and Teh Exodus Akwa^{2*} 

¹Department of Plant Sciences, Kenyatta University, Kenya

²Department of Biochemistry, Microbiology and Biotechnology, Kenyatta University, Kenya



Abstract

In Kenya, maize is a staple food and is stored after harvest as a source of food and for sale. From the stored maize, the surplus from that meant for food is sold by some farmers to earn income. The quality of the maize in store depends on the storage facility. Poorly constructed and unmaintained structures may allow leakage of water and entry by rodents and insects which may mechanically damage the maize by nibbling and holing respectively, thus predisposing the grains to fungal infection. Some of these fungi can contaminate the maize with mycotoxins. The storage fungi on maize in Kenya and the probable mycotoxin their produce were reviewed for the period of 2000 to 2015 (15-years). This review established that the most frequently isolated maize storage fungi belonged to the genera *Aspergillus* followed by *Fusarium* and lastly *Penicillium*. Further it was established that the most frequently reported mycotoxin was aflatoxin which was most prevalent in the Eastern region of Kenya. Fumonisin was also reported but had the highest occurrence in the Western region. The variation in the occurrence of these toxins could be due the differences in the agro ecological zones of the regions. It was further noted that although records indicated that other species of storage fungi were present in maize their respective mycotoxins were not established. Hence it is mandatory to carry out a complete census of storage fungi of maize in Kenya and establish whether these fungi produce the expected respective mycotoxins. This is paramount because of the health implications associated with the mycotoxin ingestion.

Keywords

Fungal Toxin, Review, Storage, Zea mays

Introduction

Maize is a cereal crop grown in most tropical countries including Kenya for human consumption and animal feed. It is the most widely grown staple food crop in Africa [1]. It is the most important staple food for the majority of the Kenyan population [2]. This is probably because it is suited to wide variety of agroecological zones and can be grown in quantities enough to last through the off growing season after harvest. It is consumed at an average intake of about 400 gm per person per day [3] Shepherd, 2008. It is used to make maize meal which is a staple food consumed by nearly all the communities in Kenya. In addition to being a source of food, it is a source of income to farmers. Furthermore, it is used to make local brew commonly known as 'busaa' which is an "opaque beer" used in the country by some communities for ceremonies or for leisure.

In Kenya when maize is harvested it is stored in varied structures by different communities. Such structures include wooden granaries, grass lined granaries and silos. In some of these structures the maize is exposed to elements that bring about infection by fungi which may lead to contamination by mycotoxins. Some of these predisposing elements include

water from leaking roofs which increases the moisture contents of the maize creating a hotspot for the growth of fungi leading to infection of the grains. Infestation by insects such as weevils bore holes on the grains thereby creating avenues through which fungi infects the grains. Some of these storage structures are not very secure and therefore provide openings through which small mammals such as rats, squirrels and mice can gain entry to the store thereby damaging the maize by nibbling. In addition, they deposit their droppings and urine which moistens the grains thus creating conditions that encourages the growth of fungi especially on grains which may have been damaged by the nibbling of the mammals.

***Corresponding author:** Teh Exodus Akwa, Department of Biochemistry, Microbiology and Biotechnology, Kenyatta University, Nairobi 43844-00100, Kenya, Tel: +254-792-775729

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Fungal infection on the stored maize leads to deterioration in quality (colour, taste, and flavour) and quantity (which constitutes post-harvest loss of maize).

In Kenya, the most frequently isolated storage fungi in maize are the *Aspergilli* which are associated with aflatoxins which cause aflatoxicosis. However, literature records indicate that stored maize is not only infected by *Aspergillus*. It is also infected by other genera of fungi particularly *Penicillium*, *Fusarium* and *Alternaria*.

This article reviews the storage fungi of maize in Kenya spanning through a period of 15-years (2000-2015). Thus relevant literature pertaining to the study was examined and the reported storage fungi on maize in Kenya was documented in a table indicating the region where the occurrence of the fungus was established, the identity of the fungus, the probable mycotoxin they produce, the associated mycotoxicosis and finally the article reference. The information documented in the table will reveal which other fungi apart from *Aspergillus* affects maize and therefore the necessity to determine the other mycotoxins that may be present on the stored maize which might probably be the cause of some of the prevalent diseases in the country such as nephropathy and oesophageal encephalopathy.

Maize Harvesting, Transport and Storage Structures in Kenya

The harvesting methods; means of transport and type containers; the kind of storage facilities and the state, in which they are, influence the quality of the maize that is in store.

Maize harvesting methods

Maize may be harvested manually or using mechanical means. Depending on the handling, each method predisposes maize to fungal infection. Manual harvesting involves de-husking the cob and throwing it to an identified site within the farm. The site may be bare or covered by the farm weeds. The de-husked cobs are normally thrown to the sites where on landing, can come in contact with soil or brush against weeds. The soil is an environment that is very rich in fungi, some of which may be capable of infecting the maize grains. Likewise, the weeds may host fungi which may also infect the maize.

Alternatively, mechanical harvesting may create damage sites on the maize, which become the foci of infection by fungi. The conditions at harvest may also predispose the maize to infection by fungi. Maize harvested during wet conditions is prone to fungal infection because moisture promotes their growth on the maize. The maturity stage at which maize is harvested is also important with respect to predisposition of maize to infection by fungi. Maize harvested when the grains are still soft; contains high moisture content; and hence easily infected by the fungi. Similarly, maize harvested late may be exposed to damage by birds and small mammals, which damage the produce by de-husking of the top part of the cob followed by feeding on the grains. When it rains, water seeps in, moistens the damaged grains, which become sites of pro-

liferic growth of fungi. The infection of the grain thus spread from the de-husked, top part of the cob, downwards. In some varieties the cob opens at the top, thus de-husking in a similar manner as the ones damaged by the animals thus getting infected by to fungi as previously described.

Transport means and containers used

In Kenya, harvested maize may be transported in gunny bags: carried manually, placed in manually pulled carts or on the backs of donkeys. Maize handled this way is rarely infected by fungi because there are not damaged. Maize handled this way arrives in stores in a state that would not be prone to the development of fungi, because the handling procedure does not inflict damage on the produce. On the other hand, in Kenya, maize is also transported in tractor pulled trailers, pickups and Lorries. Movement of the transport vehicle through a rough terrain, poor roads causes the vehicle to sway and vibrates, thus making the maize being transported to hit against the sides of the lorry and against themselves thus getting mechanically damaged by cracking or splitting thus creating sites for fungal infection.

Maize storage structures

The storage facilities are important in preserving the quality of the produce. Stores that are spacious, well aerated and without leaking roofs ensures post-harvest longevity of the produce. On the other hand, poorly constructed and unmaintained stores allow access of small mammals that nibble and pests that hole the maize grains.

Storage facilities used by farmers depend on the scale of farming. Large scale farmers use storage structures such as sophisticated silos. On the other hand, small scale farmers use all kinds of containers or bags. In Kenya, the commonly used maize storage structures by farmers include traditional wooden granaries, grass line granaries and gunny bags (Figure 1). Figure 1a shows that the maize is stored on the ground. The storage structure in Figure 1b is a grass lined wall and a grass thatched roof. The storage structure shown in Figure 1c consists of wood with an iron sheet roof. In Figure 1d the store for maize is a structure that is made of fibre. Maize storage in these structures is vulnerable to entry of water and rodents. The rodents mechanically damage the grains by nibbling on them as they feed thus creates ports of entry by fungi. The growth of fungi is promoted by the moisture arising from leaking roofs and walls as well as from urine and droppings from the rodents.

Diversity of post-harvest fungi in maize

The conditions at which the maize grains arrive at the store determine the fungi that predominate and the subsequent infection by the storage fungi. If the grains were harvested immaturity or under wet conditions, they are infected by the field fungi such as *Rhizopus*, *Mucor*, *Rhizoctonia*, *Cladosporium*, *Trichothecium*, *Fusarium* and *Altenaria*. Post-harvest fungi infections usually occur after harvest either during transit or during storage of grains. Fungi that develop on grains during storage usually survive at low moisture contents. Major examples of these storage fungi include *Asper-*



Figure 1: Different maize storage structures and practices (a) Maize cobs placed on floor; (b) Grass granary; (c) Wooden granary; (d) Storage (sisal) bag. Source: Kenya Agricultural Research and Livestock Organisation).

gillus and *Penicillium* [4]. Studies done by Lamboni, et al. [5] of Togo on traditional African granaries identified species of *Aspergillus*, *Rhizoctonia*, *Penicillium* and *Fusarium oxysporum* as post-harvest fungi commonly infecting maize in the granaries. Similar reports by Julian, et al. 1995 [6] in Honduras indicated that *Fusarium moniliforme*, *Fusarium moniliforme var. subglutinans*, *Penicillium spp*, *Stenocarpella maydis*, *Sordaria macrospora* and *Acremonium spp* were the predominant species detected in stored maize.

Mycotoxin production by storage fungi on maize

Fungi that grow on maize grains in the store not only damage and spoil the quality of maize, but they also contaminate them with toxic metabolites called mycotoxins. These mycotoxins are toxic to both animals and humans upon consumption. The mycotoxins released by fungi are the most potent metabolites known to man because they are capable of causing diseases at low concentration [7]. Two reasons have been brought forth explaining the potency of mycotoxins released by fungi. Firstly, these mycotoxins are heat resistant and

cannot be destroyed by conventional heat treatments even when the infected seeds are cooked. Secondly, their release from the fungus colony to food is rapid and affects variety of processed and unprocessed food products. For example, it can contaminate maize grains in stores; flour prepared from maize and cooked meal prepared from maize flour normally called 'Ugali'. Furthermore, brewed products such as the local alcoholic drink called 'busaa' in Kenya can also be contaminated with the mycotoxins. More interestingly, when dairy cattle are fed with mouldy grains another metabolite of the mycotoxin that is found in the grains is secreted in the milk and is as potent as the original metabolite.

Studies have shown that the most important species of fungi and mycotoxins that could contaminate maize grains include *Aspergillus flavus* releasing aflatoxins, *Fusarium verticillioides*, *Fusarium proliferatum* releasing fumonisins and *Fusarium graminearum* releasing trichothecenes and zearalenone [8,9]. Detected aflatoxin B1 (AFB1), zearalenone (ZON), deoxynivalenol (DON) and fumonisin B1 (FB1) as mycotoxins on stored maize grain samples in the institute of animal hus-

bandry, in Belgrade. Adetunji, et al. [10] also identified Aflatoxin B1 and Fumonisin B1 as mycotoxins in stored maize in five major agro ecological zones in Nigeria released by fungal species and *Aspergillus parasiticus*, and *Fusarium oxysporum* respectively. Some of the health impacts following the consumption of these mycotoxins may include liver cancer caused by consuming aflatoxins, abortion in humans following intake of zearalenone and oesophageal cancer caused by consumption of fumonisins [11].

On the other hand, reports from stored maize contami-

nation in Kenya spanning a period of over 15-years show that Aflatoxins and fumonisins are the major mycotoxins found in stored maize (Table 1) [12-25]. This is probably due to the fact that most research on mycotoxin contamination on stored maize in Kenya is focused mostly on the presence of Aflatoxins and fumonisins only. Thus, more research work has to be carried out to determine the presence of other mycotoxins in stored maize in Kenya. These other mycotoxins might pose health issues to human and livestock if research is not done to find out what they are so as to alleviate the problem.

Table 1: Fungi contamination of stored maize in Kenya.

| Year | Region/Areas | Fungal species Isolated | Mycotoxin | Mycotoxicosis | Reference (s) |
|------|--|--|------------------------|---------------|---|
| 2001 | Western Region | <i>Fusarium verticillioides</i> , <i>F. graminearum</i> , and <i>F. Subglutina</i> | Fumonisin | - | Alakonya, et al. 2009 [12] |
| | | <i>Aspergillus flavus</i> | Aflatoxin | - | |
| 2002 | Western Region | <i>Fusarium verticillioides</i> , <i>F. graminearum</i> , and <i>F. Subglutina</i> | Fumonisin | - | Alakonya, et al. 2009 [12] |
| | | <i>Aspergillus flavus</i> | Aflatoxin | - | |
| 2003 | Central Region | <i>Aspergillus flavus</i> | Aflatoxin | Aflatoxicosis | Onsongo, et al. 2004 [13] |
| 2004 | Eastern and Central Kenya | <i>Aspergillus flavus</i> | Aflatoxin | Aflatoxicosis | Lewis, et al. 2005. [14] |
| 2005 | Eastern and Coastal region | <i>A. flavus</i> S, <i>A. flavus</i> L | Aflatoxin | Aflatoxicosis | Probst, et al. 2010 [15] |
| 2006 | Coastal region Eastern region | <i>A. flavus</i> L | Aflatoxin | Aflatoxicosis | Probst, et al. 2010 [15] |
| 2007 | Eastern region, Rift valley region | <i>A. flavus</i> , <i>A. niger</i> , <i>A. terreus</i> , <i>A. versicolor</i> , <i>Fusarium</i> , <i>Penicillium</i> | Aflatoxin | Aflatoxicosis | Muthomi, et al. 2009 [16] Neyole, et al. 2008 [17] |
| 2008 | Eastern region | <i>A. flavus</i> , <i>A. niger</i> , <i>A. terreus</i> , <i>A. versicolor</i> , <i>Fusarium</i> , <i>Penicillium</i> | Aflatoxin | Aflatoxicosis | Muthomi, et al. 2009 [16] |
| 2009 | Eastern region | <i>A. flavus</i> , <i>A. niger</i> , <i>A. terreus</i> , <i>A. versicolor</i> , <i>Fusarium</i> , <i>Penicillium</i> | Aflatoxin | Aflatoxicosis | Muthomi, et al. 2009 [16] |
| | Western region, Rift valley region, Nyanza | <i>Aspergillus</i> sp <i>Fusarium</i> spp | Aflatoxin Fumonisin | - - | Mutiga, et al. 2015 [18] |
| 2010 | Eastern region, Coastal region | <i>Aspergillus</i> spp, <i>Fusarium</i> spp, <i>Penicillium</i> spp, | Aflatoxin | Aflatoxicosis | Gathogo, 2010 [19] Nyaga, 2010 [20] Okoth, et al. 2012 [21] |
| | Rift valley region | <i>Aspergillus flavus</i> , <i>Aspergillus parasiticus</i> | Aflatoxin | - | Okoth et al. 2012 [21] Muthomi, et al. 2010 [22] |
| 2011 | Eastern and South Western region | <i>Aspergillus</i> spp | Aflatoxin | - | Mahuku, et al. 2019 [23] |
| 2012 | - | - | - | - | - |
| 2013 | Eastern region | <i>Aspergillus</i> spp | Aflatoxin | - | Sheila, et al. 2017 |
| 2014 | Eastern region | <i>Aspergillus flavus</i> L strain, <i>A flavus</i> S A. <i>parasiticus</i> , | Aflatoxin | - | Eliphus, 2014 [24] |
| | | <i>Fusarium. Verticillioides</i> , <i>F. proliferatum</i> and <i>F. Oxysporum</i> | Fumonisin | - | |
| 2015 | Eastern region | <i>Aspergillus flavus</i> strain L, <i>A flavus</i> , <i>A niger</i> , <i>A. parasiticus</i> | Aflatoxin | - | Maina, et al. 2016 [25] |

Discussion

In Kenya maize is a cereal that is widely grown throughout the country and therefore yield depends on the agro-ecological environments where the crop is located. Some farmers produce maize in excess of what they need for food. The surplus is stored until the market prices are best then sold as a source of income that is desperately needed by the poor resource rural farmers to improve their livelihoods. The quality of maize at the time of selling depends on the duration of storage, the quality of the store and the state of the maize before storage. The quality of the maize may progressively deteriorate over the storage period due to damage by rodents which may gain entry if the stores are not secure. For example, the maize placed on the floor (Figure 1a) are easily accessed by rodents because there are not protected by any structures. Figure 1b shows the store with a grass thatched roof. Such stores may allow the leakage of rainwater into the store therefore moistening the grains thus predisposing them to fungi infections and subsequently spoilage. Likewise, the wooden stores (Figure 1c) when aged may allow rodents to get in and therefore spoil the maize. Although the storage bag (Figure 1d) seems to be well sealed, the sides is likely to be destroyed by rats because there are capable of biting and breaking the fibres used in making the bags and therefore gain access into the bag and consequently damaging the maize as there feed on the grains thereby predisposing the grains to fungi infection. Furthermore, the maize can also be damaged by insect pests if they are not controlled. Apart from discoloration of the grains resulting from the invasion by the fungi the flavour and the taste of the grains changes. This therefore lowers the quality of the maize and the excessively damaged ones may be discarded while the moderately damaged ones will fetch a lower price in the market compared to the clean grains. Apart from lowering the quality due to the moulding of the maize by the storage fungi some of these fungi may release mycotoxins. The kind of mycotoxin depends on the fungal species. According to the results obtained from this study (Table 1) [12-25] it was established that three genera (*Fusarium*, *Aspergillus* and *Penicillium*) were found to infect the maize during storage. Amongst which *Aspergillus* was the most frequently reported. The presence of these genera of fungi are probably indication that the maize may be contaminated by the respective mycotoxins there produce.

From the data which is recorded (Table 1) it is established that Eastern Kenya was the region in which *Aspergillus* was the most frequently encountered genus present in stored maize and this may explain why aflatoxins was most frequently detected in the maize in this region (in 12 out of the 15-year period; 2000 to 2015). Eastern Kenya has a relative humidity of 60%-70% and temperature values of 23 °C - 34 °C [26]. Studies done by Shehu and Bello, et al. [27] showed that *Aspergillus* spp grows best above 55% relative humidity. This growth condition (above 55% relative humidity level) is comparable to the prevailing condition (60% - 70% relative humidity) in Eastern Kenya. Also, Studies done by Asevedo, et al. [28] in Brazil on the influence of temperature on production of aflatoxins in samples of stored maize artificially inoculated with *Aspergillus flavus* indicated that maximum levels aflatoxin

production occurred at 25 °C on stored maize. This temperature for the maximum growth of aflatoxin is also comparable to the prevailing condition of Eastern Kenya (23 °C - 34 °C). The humidity levels and temperatures cited as prevailing conditions of Eastern Kenya probably explain why Eastern Kenya is the region where *Aspergillus* and aflatoxin contamination is reported to be most frequently encountered on stored maize in Kenya compared to the other regions as evident in Table 1. This may also further confirm why aflatoxin was the most frequently observed mycotoxin on stored maize in this region (in 12 of the years out of the 15-year period; 2000 to 2015). In the same period of the study it was established that Fumonisin, a mycotoxin that is produced by the genus *Fusarium* was found to contaminate maize in Western Kenya (2001, 2002) [12,16,24]. Whereas in 2014 it was reported in Eastern Kenya [24]. The record showed that Fumonisin was recorded in four of the years over the entire study period. In which in 3 of these years it was observed in Western Kenya and in the remaining year in Eastern Kenya. The probable reasons for the higher occurrence of Fumonisin. Western, than in Eastern Kenya could be explained by differences in the prevailing environmental conditions. Western region of Kenya is wetter and cooler than the Eastern region. This environment that is suitable for the growth of maize, is also suitable for the development for the *Fusarium* that releases fumonisins. The occurrence of fumonisins on maize in Eastern Kenya in 2014 could have been due to abnormally wet conditions due to high rains which created conditions suitable for the growth of fumonisins on maize which may explains why fumonisins were isolated in this year only. On the other hand the maize samples which were found to contain fumonisins may have originated elsewhere as commercially sold to the consumers in Eastern Kenya or they might have been the government relief food which is normally given to the residents of that area because drought and famine is a common occurrence in Eastern Kenya. Furthermore, it was observed that in rift valley, a region where the greatest amount of maize is produced in Kenya, the presence of *Fusarium* was only reported twice (2007 and 2009) and even then, no fumonisin was reported. Although it is expected that fumonisin toxins should be present on maize yearly, the information established over the study period did not conform to this expectation. It was established that fumonisins were present on maize in only three of the years (2001, 2002 and 2009) spanning through the 15-year period. The observed data depicts that: There is no consistency in monitoring of the possible presence of the mycotoxin in stored maize yearly. It is possible that there might be lack of awareness that Western Kenya is an environment which is suitable for the growth of the *Fusarium moniliforme* and possible its presence on stored maize hence no concern about the possible presence of that contaminant. *Aspergillus aflatoxins* may have over shadowed other mycotoxins that are found on maize because it was the most frequently encountered mycotoxins that was found on food products and is linked to a serious ailment in humans notably liver cirrhosis. This could be why more research effort is directed to the study of aflatoxins unlike all the other mycotoxins including fumonisins which is also known to have been linked to a very serious disease (oesophageal cancer) which is one of the most serious and

prevalent disease in the country. In addition, published information indicates that the source of the two toxins; *Aspergillus* for aflatoxins and *Fusaria* for Fumonisin are not the only Fungi found in stored maize. There are others such as *Penicillium* which is known to produce ochratoxin that is known to cause kidney nephropathy and urinary tract tumours [29-39].

Conclusion

Maize is a staple food and a major source of income to the poor resource farmers. The nature of the storage structures used by farmers to store maize determines the preservation of the quality of the maize during the storage period. The most important agent of spoilage maize in the store is the presence of storage fungi. From the study it was established that fungi of the genera *Aspergillus*, *Penicillium* and *Fusarium* were responsible for the spoilage of the maize grains in store. With respect to the distribution of the storage fungi in the country, *Aspergillus* was established to be the most encountered fungus in the Eastern region. While *Fusarium* was the most encountered in the Western region. Similarly, the distribution of the mycotoxin was dependent on the predominance of the genera in the respective region. With respect to mycotoxin distribution, aflatoxin was most frequently encountered in the Eastern region while Fumonisin was the most encountered in the Western region. This was in conformity with the distribution of the respective fungi. The predominance of the fungi in the respective region is probably determined by the prevailing agro climatic conditions in the region. Likewise, too the distribution of the mycotoxin in relation with the respective genera of fungi.

From the study, it was established that there are more reports on the presence of aflatoxins in maize in Kenya. It was also established that the frequency of reports of fumonisin was far much less compared to aflatoxins. Additionally, in areas where *Fusarium* was recorded for example Coastal region and rift valley fumonisin was not recorded. It is therefore not known whether *Fusarium* which infects maize in stores in these areas produces fumonisin. Yet it is expected that because rift valley is the most suitable region for growing maize, it should also be possible for *Fusaria* to attack maize and contaminate with *fumonisin* but the information recorded does not show. The absence of the record of fumonisin in rift valley may indicate that there has been no research to fumonisin contamination of maize in the area.

Additionally, *Penicillium* was one of the fungi that was found to infect maize in store but there is no report on the presence of its expected mycotoxin.

Recommendations

The policy makers should advice farmers to construct storage structures which ensures that there is enough air circulation to dry the maize, secured enough to prevent rodents from accessing the stores and roofs safe enough not to allow leakage of rain water. In addition to that, farmers should control the entry of insect pests in stores to avoid the damage there cause on the grains.

A complete census of the storage fungi of maize should be carried out in Kenya and also establish whether these fungi

produce the expected respective mycotoxins. Furthermore, it should be established if the levels of the different kinds of mycotoxins contaminating maize are within or above their safe limit.

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