

Review

Scientific understanding and effects on ear rot diseases in maize production: a review

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Maize ear rot disease is global phenomena that results on significant yield loss and risks of health to humans and animals. It is also leading constrain on the loss of grain yields due to its occurrence which damage yield and quality of the maize. Moreover, ear or kernel damage of the maize can be caused by a number of fungi, especial *Fusarium* species. The occurrence of ear rots can vary greatly from year to year as well as field to field. These diseases cause of great concern in maize production worldwide, as it is considered to have a severe impact on the maize production by farmers and it reduces the grain potential income in the market. Therefore, this review can provide a better understanding about effects of ear rot diseases and their implication on maize production. It is also imperative to be thoughtful about these in order to increase grain yields to meet future food requirements amid strong competition for limited resources. Besides, more attention also required on the development of maize cultivars with broad-based resistance to the pathogens. Therefore, this paper aims to understand effects of ear rot disease on maize production.

Keywords: Ear rot diseases, *Zea mays* L, *Fusarium* species

INTRODUCTION

Maize (*Zea mays* L.) is can be defined as a dicotyledonous angiosperm plant that belongs to the grass family (Poaceae) (Park, 2001). This valuable crop is cultivated worldwide and represents a staple food for a significant proportion of the world's population (Anderson et al., 2004). In China, maize plays a very significant role in grain production. Wang et al. (2005) point out that maize ranks second after rice in planting area and average annual planting area is 24 million ha with the total yield of 125 million ton. In the world, China is ranked as second largest maize producing country (Wang et al., 2005; Statista, 2014). The loss of maize productivity due to diseases is a worldwide phenomenon (Oerke, 2005). Balint-Kurti and Johal (2009) indicated that loss due to diseases were estimated as 4% from Northern Europe and 14 % from West Africa and South Asia.

There are majority of reports that report maize diseases that affect roots, stalks, ears, and kernels which caused by fungi (White, 2000). Among these diseases, ear rot is one of the most important in all the countries where this cereal is grown. The notable incidence of ear rot occurs in the moist and humid regions of southwest China, as well as other regions with similar longitude in other countries (Wen et al., 2002; Ali et al., 2005). *Gibberella* ear rot, *Fusarium* ear rot, and *Aspergillus* ear rot are three predominant ear rot diseases of maize (Xiang et al., 2010). These three diseases are responsible for most disease-related reductions in yield and quality. Ear rot diseases are related to many factors such favourable condition for *Gibberella* ear rot (Vigier et al., 2001). In China, maize ear rot is more associated to the feeding of Asian corn borer on silk and kernels (Xia el al., 1995; Wang et al.,

2005). Therefore, damage caused by Asian maize borer results on the contamination of maize grains by mycotoxins from *Fusarium* species (Wang et al., 2005). Moreover, *Fusarium* species are extensively distributed worldwide from temperate to tropical regions (Leslie and Summerell, 2006). In China, maize ear rot caused by *Fusarium* spp are leading to more significant yield losses and potential risk of mycotoxin contamination (Scauflaire et al., 2011; Wang et al., 2014 b). A better understanding of the ear rot diseases is imperative that will accelerate progress in disease prevention and management. Therefore, this paper aims to understand effects of ear rot disease on maize production.

Most common fungal diseases on maize ears

The most predominant types of maize ear rot diseases in the world has been related to *Aspergillus* ear rot, *Fusarium* ear rot and *Gibberella* ear (Xiang et al., 2010). *Aspergillus* ear rot caused by *Aspergillus flavus* (Link: Fr), while *Fusarium* ear rot is predominantly caused by *Fusarium verticillioides* (Sacc.) Nirenberg (synonym: *F. moniliforme* Sheldon; teleomorph: *Gibberella fujikuroi*) and *F. Proliferatum* (Matsushima) Nirenberg (teleomorph: *G. intermedia*). These diseases are most serious in hot and dry weather (Zuber et al., 1978; Marasas et al., 1979, Payne and Widstrom, 1992). *Aspergillus* ear rot contaminates the grain with aflatoxins, whereas *Fusarium* ear rot generally results in the accumulation of fumonisins, moniliformin and/or beauvericin (Logrieco et al., 2002). In fact, *Fusarium* species produce secondary metabolic compounds (mycotoxins) that may render the grain inedible or toxic to domesticated animals and humans at sufficient concentrations (Parsons, 2008). Munkvold and Desjardins (1997) also mentioned that *Fusarium* species are capable of causing seedling diseases, root rots, stalk rots, and ear rots of maize as well as damaging stored grain. *Gibberella* ear rot results in contamination from two main mycotoxins, deoxynivalenol and zearalenone. All of the mycotoxins described above can cause severe diseases in livestock and humans (Gelderblom et al., 1988; Castegnaro and McGregor, 1998; Pestka, 2007; Voss et al., 2007; Henry et al., 2009). In China, *Fusarium* ear rot is noted as one of most important ear rot disease that severely affect maize yield China (Ding et al., 2008; Yuan et al., 2012; Yuan et al., 2013; Wang et al., 2014 a, b). *Fusarium* species that cause maize ear rot in the different areas in China are highly diverse and areas with high levels of fumonisin contamination have a potential health risk for human and animals (Scauflaire et al., 2011; Wang et al., 2014 b; Fu et al., 2015). Study conducted by Fu et al. (2015) also revealed that higher

incidence of *Fusarium* species were reported in Guizhou province than other 8 sampled provinces. Recently, many studies conveyed a countable number of first report from *Fusarium* maize ear rot including *Fusarium kyushuense* (Wang et al., 2014 b), *Fusarium temperatum* (Zhang et al., 2014 b), *Fusarium andiyazi* (Zhang et al., 2014 a), and *Fusarium meridionale* (Zhang et al., 2014 c).

Maize resistance in relation to ear rot diseases

The most dangerous food and feed safety challenge on maize production is ear rot diseases of the *Fusarium* species. Mesterhazy et al. (2012) pointed out that most of the inbreds and hybrids are susceptible to ear rot diseases (Mesterhazy et al., 2012). Sporadically, more yield losses through *Fusarium* species such as *Fusarium graminearum* and *Fusarium verticillioides* have been reported (Vigier et al. 2001). This phenomenon has led to some sporadic breeding efforts which undertaken to increase resistance to ear rot diseases (Mesterhazy et al., 2012). Initially, researchers reported that resistance of the disease is inherited in a quantitative manner (Boling and Grogan, 1965; Ullstrup, 1977) until *Fusarium* mycotoxins were reported as main cause of the economic loss through ear rot. Gelderblom et al. (1988) reported the *Fusarium* mycotoxins of the grain as a *verticillioides* mycotoxin, fumonisin B1. The discovery of *Fusarium* mycotoxins from the grain has opened a window several years ago for increasing ear rot resistance (Mesterhazy et al., 2012). Visconti and Doko (1994) pointed out that under laboratory conditions, isolates of *F. verticillioides* and *F. proliferatum*, derived from maize, synthesise more FB 1 (average 1259 ppm) than the isolates originating from grains of wheat (average 769 ppm) or barley (average 320 ppm). Therefore, fumonisin B1 concentration depends on the origin of isolates, including a host plant and a geographic region. Moreover, host resistance offers the most economical means of controlling these ear diseases in maize. However, most maize genotypes are highly susceptible and source of good resistance are few (Reid et al., 2009).

The most promising avenue to control ear rot disease is the breeding of maize for resistance. Moreover, this approach is more applicable on the situation of less available controlling strategies. Therefore, developing resistance to ear rot is an important objective in maize breeding programs (Robertson-hoyt et al., 2007; Bolduan et al., 2010). Regrettably, Xiang et al. (2010) pointed out that there is no evidence of complete resistance to ear rot in maize.

It has been also noted that ear insect wounds from pests such as *Ostrinia nubilalis*, *Diatraea grandiosella*, *Diabrotica virgifera virgifera*, *Heliocoverpa zea* and *Frankliniella* species can significantly increase chances of *Fusarium* infection due to created new points of entry whereby the fungus usual enter the plant (Parsons and Munkvold, 2010). Therefore, resistance of the maize required to be integrated with insect resistance to maximise chances of ear rot control. Davis et al. (1989) also reported that after four decades of resistance to *Fusarium* ear rot in maize taken place so far, no lines are immune against this disease and the mechanism of resistance has not been well understood. In general, the important point is in determining resistance of cultivars to ear rot, pathway of infection of maize grain has a special importance.

Morphological and Molecular identification of *Fusarium* spp

The identification of *Fusarium* species is mainly based on distinctive characters of the shapes and sizes of macro and microconidia, presence and absence of chlamydospores as well as colony appearances, pigmentations and growth rates on agar media (Leslie and Summerell, 2006). Nevertheless, the most important step in the process of identification includes morphological identification of pathogenic fungi. This step may not suffice to complete identification which requires more analysis to be added (Summerell et al., 2003). Rahjoo et al. (2008) stated that some species cannot be reliably identified in this way, especially for members of the *G. fujikuroi* complex, where further analysis such as DNA sequencing and species-specific PCR assays must be conducted.

Moreover, it has been further noted that differentiating species within the *G. fujikuroi* species complex using morphological characters is difficult even for specialists (Summerell et al., 2003; Leslie and Summerell, 2006). Hence, DNA sequence-based identifications and species-specific PCR assays are usually needed to accurately identify species within the complex and some cases these analysis are used to confirm morphological identifications and to identify unknown isolates (Rahjoo et al., 2008).

Vitale *et al.* (2011) also pointed out that it is commonly accepted to be difficult to identify members of the genus *Fusarium* at the species level if simply relying on morphological traits. Moreover, morphological identification requires considerable expertise, especially in distinguishing closely related *Fusarium* species, as their morphological features may overlap (Sever et al., 2012). Therefore, molecular approaches will support

morphological diagnostics via providing a rapid and reliable assay for routine identification *Fusarium* species.

Maize ear rot disease in relation to yield and quality

Maize is the host for a large number of pathogens, which invade all of its organs from the germination until the harvest, ear and grain infection often remain even during the storage (Nagy et al., 2006). Fungal entry into maize ears after silking occurs through two major modes: (a) by growth of mycelium down silks to the kernels and cob (rachis) from spores germinating on the silks, and (b) by entry through kernel wounds, caused by insects, birds and hail (Hesseltine and Bothast, 1977, Reid et al., 1996). Therefore, it leads to ear rot diseases to have a direct effect on maize kernels (Yuan et al., 2013). Ultimately, ear and kernel rot diseases decreases the grain yield quality and feeding value of the grain, and in some instances, this has even resulted in the production of toxic substances in feed rations (Bello et al., 2012).

Xiang et al. (2010) also mentioned three predominant ear rot diseases of maize such as *Aspergillus* ear rot, *Fusarium* ear rot and *Gibberella* ear rot. These three diseases are responsible for most disease-related reductions in yield and quality. Ear rot diseases can directly cause grain yield losses. Once infection occurs from ears or kernels, it affect appearance and quality of kernels (Reid et al., 1996; Vigier et al., 2001; Yuan et al., 2013). In a favourable environment, moderately severe *Gibberella* ear rot symptoms can result in up to 48% yield reduction (Vigier et al., 2001). Moreover, ear rot diseases of maize through their symptoms reduce significantly the quantity and the quality of the yield, estimated between 7-17% but, in the favorable years for the diseases, they can be much larger (Nagy et al., 2006).

Ear rot disease results in reduced grain but the main loss from ear rot disease is due to the contamination of grain yield with mycotoxins which are a threat to safety of both humans and livestock (Voss et al., 2007; Pestka, 2008; Bello et al., 2012). Ear and kernel infection by these fungi is of great economic importance quality due to grain rot and discolouration which lead to yield and quality loss (Richards, 2007). The quest for improved grain yield and disease tolerance/ resistance maize varieties therefore become imperative for profitable maize production.

Symptoms of ear rot diseases in Maize (*Zea mays* L.)

Symptoms of maize ear rot diseases varies a lot depending on the causal agent (fungi), maize genotype,

and disease severity from region to region. Wang et al. (2014 b) reported that clear symptoms of *Fusarium* ear rot is white to pink or salmon-colored mold at the tip of the ear. Among maize ear rot, *Gibberella* ear rot cause by *F. graminearum* has been reported as it shows a pinkish coloured mould (White, 1999). In fact, the major symptom of *F. graminearum* infection on maize ears is a characterised by pink to reddish coloured mould on kernels and between husks (Reid et al., 1996). Generally, this disease begins as white mycelium moving down from the ear tip whereby it later turns reddish-pink on infected kernels. This phenomenon varies a lot due to the fact that sometimes pinkish fungal growth can be found on the exterior husk leaves, and in severe infections (Reid et al., 1996), it is impossible to separate the husks from the kernels as the entire ear becomes a tightly bound mass of fungal (Mesterhazy et al., 2012). The developmental rate of the symptoms from year to year is quite dependent on the environmental factors that can influence, not only ear development but subsequent kernel dry down and also fungal growth. However, Christensen and Kaufmann (1969) pointed out that from the range of 22-23% moisture reached by kernels, it tend to be less for the fungus to proceed its infection. Mesterhazy et al. (2012) indicate that there is a situation where cob is only part infected and no symptoms visible to the ear but a cob hand squeeze will lead to a wet cob with pink or reddish colour.

In contrast, *Fusarium* ear rot of maize is commonly known as it is caused by *Fusarium verticillioides*. Therefore, *Fusarium* ear rot from *F. verticillioides* is often associated with insect infestations or maize earworms (Mesterhazy et al., 2012; Dragich and Nelson, 2014). *F. proliferatum* and *F. subglutinans* are also minor causal agents of *Fusarium* ear rot, as are probably other members of the Liseola Section of *Fusaria*. (Iglesias et al., 2010). The symptoms of this disease occur mainly on individual kernels or on limited areas of the ear (White, 1999). The distinct symptoms of *Fusarium* ear rot is white, pale pink, or pale lavender mycelia (Dragich and Nelson, 2014). Ncube (2012) mentioned that individual or groups of infected kernels are usually scattered randomly on the entire ear and they appear as whitish and pink kernels. In the most cases, this disease tend not to cover the whole ear, instead; it remains around injured regions caused during feeding (Dragich and Nelson, 2014) but on the severe cases may manifest as scattered areas of infection increase over the ear surface (Ncube, 2012). Therefore, it becomes imperative to understanding these symptoms and signs that will lead to an easy visual diagnosis of the diseases with less effort and cost required.

Climatic conditions in relation to ear rot disease

The increase of maize ear rot diseases can be associated with climatic conditions (temperature and rain-fall), although many different pathogens occurs in maize grain. It has been noted that maize is predominantly infected by *Fusarium* species such as *F. graminearum*, *F. moniliforme*, *F. proliferatum* and *F. subglutinans* (Doohan et al., 2003). These *Fusarium* species vary with the climatic distribution and their optimum climatic conditions for their persistence. Several *Fusarium* species can affect a single ear or kernel (grain) depending on the climatic growing season of the region (Logrieco et al., 2002 b).

Fusarium diseases are complicated by the fact that *Fusarium* fungi can cause disease individual or in complex infections (Doohan et al., 1998). In fact, these fungi rarely exist in isolation, but occur as a complex with each other and with other *Fusaria* and other fungal genera (Doohan et al., 2003). Moreover, these species respond differently to the environmental variations, particularly temperature and osmotic stress (Conrath et al., 2002). Therefore, climate influence competition between, and the predominance of different fungi within this complex.

Climatic conditions during harvesting/storage time may influence contamination of maize ears (by toxigenic fungi and mycotoxins) until it is consumed by humans and animals. Due to the fact that, climatic conditions play an important role in the growth of *F. verticillioides* which accumulates fumonisin prior and post-harvest (Miller, 2001; Ono et al., 2002). Sweets and Wright (2008) mentioned one exception about *Aspergillus flavus*, which favoured by drought stress to maize during pollination and warm temperatures as kernels matures. *Fusarium* ear rot occurs in all areas where maize is grown because of the pathogens which can live within the maize plant without causing apparent disease (Ncube, 2012). *Fusarium* ear is most severe under hot, dry weather conditions that occur after flowering. Moreover, Sweets and Wright (2008) point out that most of the fungi are more prevalent when the rainfall is above normal from silking to harvest. The influence of climatic condition on the incidence of *Fusarium* species is probably both direct (an effect on mode of reproduction) and indirect (an effect of soil and vegetation type). Doohan et al. (2003) suggested more research to determine the indirect effect of climate on the incidence of *Fusarium* fungi and how this affects species-specific factors.

Use of inoculations methods and time of inoculation

The inconsistency of natural infection from year to year has triggered maize researchers to use artificial inoculation methods to inoculate the plant material with fungal spores (Schaafsma et al., 1997). Therefore, use of inoculations methods became a centre of breeding programs whereby a number of artificial inoculation methods and their variants have been developed (Mesterhazy et al., 2012). The most common used methods involves use of suspension to inject it into the silk channel or the centre of the ear via the syringes (Reid et al., 1996). The use of silk channel alone is not sufficient due to the fact that infections from the kernels can occur at several stages of ear development. Also, kernel channel alone may not be enough because infection occurs on the silk while kernels are not yet matured which can lead to the severe infection of kernels with cobs (Reid and Hamilton, 1996).

Reid et al. (1996) described these inoculation methods as follow; screening for silk resistance has usually involved one of three techniques: (1) insertion of a colonized substrate (e.g. toothpicks or cereal kernels overgrown with mycelium) or a pipe cleaner impregnated with macroconidia into the silk channel (region within the husk between the tip of the cob and tip of the husk where the silks emerge); (2) spraying a conidial suspension on the exposed silks; or (3) injection of a conidial suspension into the silk channel. Then, screening for kernel or wound resistance often involves puncturing the husk, kernels, and cob followed by insertion of a colonized substrate (toothpick) or spores (saturated pipe cleaner) into the wound. However, Mesterhazy et al. (2012) pointed out that these methods can be used for artificial inoculation and the evaluation of resistance for *Fusarium* ear rot, other *Fusarium* species as well *Gibberella* ear rot. Reid et al. (1992) reported that injection of a conidial suspension into the silk channel gave consistent results and allowed for differentiation between resistant and susceptible genotypes. Moreover, Chungu et al. (1996) compared numerous inoculation methods and concluded that silk channel inoculation was effective in measuring silk resistance.

Several inoculation methods were also compared in the study conducted by (Clements et al., 2003) where inoculum was injected through the ear husk, inoculation with different variants were sprayed and six *Fusarium*-colonized tooth-picks were inserted into the silk channel. Among all evaluated methods, only the injection through the husk leaves significantly increased fumonisin concentration and infection severity. Also, Eller et al. (2008) compared several inoculation methods but noted that the highest infection severity and largest genotypic

differentiation on the inoculum inserted through the husks. However, Bush et al. (2004) tested several methods and concluded that the best method was penetrating husks with pin bars and injecting inoculum down the silk channel.

Time of the inoculation is one of the most important factor during inoculation. Reid et al. (2002) stated that timing can be based on the number of days from 50% silk emergence (50% of the plants of a given genotype with emerged silk) and silk channel inoculations are best done 4-7 days after silk emergence when there is a peak in expression of susceptibility. Moreover, Schaafsma et al. (1997) concluded that the beginning of silk browning is the ideal time for the silk channel inoculation. For kernel resistance, the best differentiation of genotypes was achieved when the kernel inoculation was performed 15 days after mid-silking (Reid and Hamilton, 1996, Reid et al., 2002). The differences in times for silk versus kernel inoculations reflect the more rapid senescence of the silks than the dry down of the kernels (Reid et al., 2002). Mesterhazy et al. (2012) further explain that adding 1 week (5–7) days post silking of the silk channel inoculations can lead to achieve the optimum time for inoculating the kernels at the same stage in which the fungus growing down the silk would reach the kernels.

CONCLUSION AND RECOMMENDATION

Ear rot is a destructive disease of maize worldwide due to the fact that it occurs widespread in maize producing areas. It has been noted that *Fusarium* species can be the predominant pathogens causing maize ear rot, a disease that results in severe economic losses and serves as a potential health risk for humans and animals. Therefore, this review can provide sufficient information which will lead to development of management practices, and therefore improve maize production in the affected areas. Also, exploration and proper disease identification will be important to help to understand more about the diseases prior to the intervention. Nevertheless, studies on effects of ear rot on maize at smallholder farmers are required to address the issue of this disease and advisory plans to prevent ear rot diseases will be beneficiary to smallholder farmers. Lastly, further research that evaluates resistance of the cultivars is also imperative.

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