Comprehensive REVIEWS in Food Science and Food Safety

Pre- and Postharvest Strategies to Minimize Mycotoxin Contamination in the Rice Food Chain

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Abstract: Rice is part of many people's diet around the world, being the main energy source in some regions. Although fewer reports exist on the occurrence of mycotoxins in rice compared to other cereals, fungal contamination and the associated production of toxic metabolites, even at lower occurrence levels compared to other crops, are of concern because of the high consumption of rice in many countries. Due to the diversity of fungi that may contaminate the rice food chain, the co-occurrence of mycotoxins is frequent. Specific strategies to overcome these problems may be applied at the preharvest part of the crop chain, while assuring good practices at harvest and postharvest stages, since different fungi may find suitable conditions to grow at the various stages of the production chain. Therefore, the aim of this review is to present the state-of-the-art knowledge on such strategies in an integrated way, from the field to the final products, to reduce mycotoxin contamination in rice.

Keywords: mycotoxin contamination, mycotoxin mixtures, postharvest strategies, preharvest strategies, rice

The Rice Plant

Rice (*Oryza sativa* L.) is one of the most important cultivated grain crops and is the staple food of nearly half of the world's population (Chen, Son, & Chang, 2012). Approximately 470 million metric tons of rice was produced worldwide in 2016 and about one-third of it in China (U.S. Department of Agriculture [USDA], 2017) (Figure 1).

Growth and development of rice from seed begin with germination and end with the formation of grain. During this period, growth and development of the rice plant can be described using a *Biologische Bundesanstalt, Bundessortenamt und Chemische Industrie* (BBCH) scale (Lancashire et al., 1991), but it can be simply divided into three principal stages: (i) vegetative, from germination (BBCH 00) to panicle initiation (BBCH 30); (ii) reproductive, from panicle formation (BBCH 32) to flowering (BBCH 61); and (iii) ripening, occurring from end of flowering (BBCH 69) to the mature grain (BBCH 89).

Botanically, rice is a caryopsis consisting of a loose husk enclosing a kernel. The kernel is made up of three parts: the outer layer includes the pericarp (seed coat) with an underlying aleurone layer, starchy endosperm, and the germ or embryo. Endosperm is full of starch and represents 90% of the kernel, the rest is formed by aleuronic layer and germ, collectively called bran, and it is rich in other nutrients like fats, proteins, vitamins, and minerals (Singh & Sinha, 2013).

The rice is commonly called "paddy" when harvested because the grain is still covered by the husk. After husk separation, the rice is called "brown" and it can be passed through polishing and sorting processes before being called "white," which is the most used form for food (Figure 2).

Rice is very sensitive to temperature during all its growth stages. Most of the rice production occurs in regions where temperatures are suitable for crop growth (day-time maximum 28 °C and night-time minimum 22 °C) (Krishnan, Ramakrishnan, Reddy, & Reddy, 2011). It is estimated that each 1 °C increase in the day-time maximum and the night-time minimum temperatures, within the 34 °C to 27 °C, and to 28 °C to 21 °C ranges, can decrease rice yield by about 7% to 8% (Baker, Allen, & Boote, 1992). For this reason, ongoing climate change can pose a risk to rice productivity and availability due to possible increases of abiotic and biotic stresses (Intergovernmental Panel on Climate Change [IPCC], 2007).

Rice grains can be affected by different diseases (Webster & Gunnell, 1992). Kushiro (2015) identified 24 kinds of unhealthy rice grains, of which 18 were moldy rice grains. Fungi may affect rice in the field and postharvest, and they contribute significantly to yield losses (Oerke & Dehne, 2004). The worldwide yield loss due to different types of pest was reported at around 37% for rice (Oerke, 2007), while the postharvest losses in developing countries accounted for 15% to 16% of basal rice production and the figure can rise to as much as 40% to 50% in countries with challenging climatic conditions (Food and Agriculture Organization [FAO], 2004). Most of these losses result from inadequate drying and unsuitable storage facilities.

CRF3-2018-0240 Submitted 10/14/2018, Accepted 12/6/2018. *Authors Gonçalves, Lima, and Venâncio are with the CEB - Centre of Biological Engineering, Univ. of Minho, 4710-057, Braga, Portugal. Authors Palumbo, Battilani, and Giorni are with the Faculty of Agriculture, Univ. Cattolica del Sacro Cuore, Via Emilia Parmense 84, 29100, Piacenza, Italy. Authors Gkrillas and Dall'Asta are with the Univ. degli studi di Parma, Via Università 12, 43121, Parma, Italy. Author Dorne is with the European Food Safety Authority (EFSA), Via Carlo Magno 1A, 43126, Parma, Italy. Direct inquiries to author Venâncio (E-mail: avenan@deb.uminho.pt).*

doi: 10.1111/1541-4337.12420

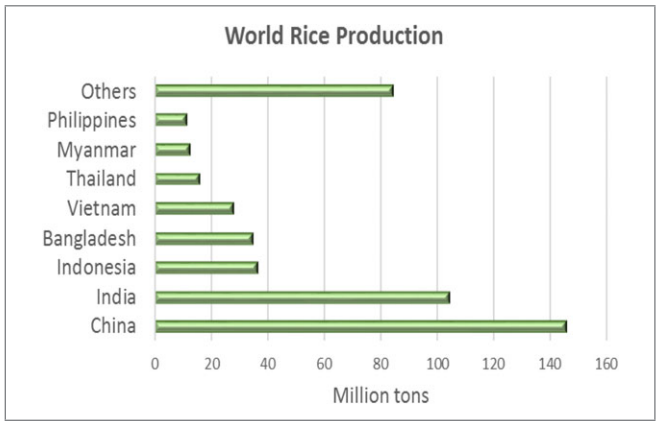

Figure 1–World rice production obtained in year 2016 (USDA, 2017).

Mycotoxin-producing fungi are listed among those able to contaminate rice and they belong to the three well-known genera *Aspergillus*, *Fusarium*, and *Penicillium* (Ferre & Santamarina, 2010; Hoeltz, Fagundes, LoboAlcayata, & Noll, 2009; Juan, Zinedine, Idrissi, & Manes, 2008; Reddy, Reddy, & Muralidharan, 2009a). Fungi can infect grains in the field and their activity can continue in the warehouse during storage, if ecological conditions remain suitable. Mycotoxin contamination depends on different factors such as the mold species, their interactions with other microorganisms, and the production area, and all are related to weather conditions and agricultural and postharvest practices (Ferre, 2016).

Although different studies exist, reports on the mycotoxin contamination of rice are fewer than those on many other cereal products (Tanaka, Sago, Zheng, Nakagawa, & Kushiro, 2007). Mycotoxin co-occurrence is also possible, increasing the toxicity of the contaminated material (Ferre, 2016).

Rice provides 20% of the world's dietary energy supply, followed by wheat (19%) and maize (5%) (FAO, 2004), and it is routinely consumed by over 4.8 billion people in 176 countries. It is the most important food crop for over 2.89 billion people in Asia, 40 million people in Africa, and 150 million people in the Americas (Otenga & Sant'Anna, 1998). Due to its high consumption rate worldwide, rice is a potentially important source of mycotoxin exposure for humans (Bhat, Rai, & Karim, 2010). Therefore, it is crucial to define the current state-of-the-art and the lack of knowledge in order to reduce consumer exposure.

Fungal Infection of Rice

Aspergillus spp. in rice and mycotoxin production

The known *Aspergillus* species that are able to produce mycotoxins in rice are reported in Table 1.

Table 1–Main fungal species reported on rice and respective mycotoxins produced.

Species	Mycotoxins
***Aspergillus* spp.**	
A. flavus	Aflatoxin B1, aflatoxin B2
A. ochraceus	Ochratoxin A
A. versicolor	Sterigmatocystin
***Fusarium* spp.**	
F. armeniacum	HT-2, T-2
F. fujikuroi	Fumonisin B1, gibberellic acid, moniliformin
F. graminearum	Deoxinivalenol
F. proliferatum	Fumonisin B1
F. subglutinans	Beauvericin, fusaproliferin, moniliformin
***Penicillium* spp.**	
P. aurantiogriseum	Penicillic acid
P. citreonigrum	Citreoviridin
P. citrinum	Citrinin
P. commune	Cyclopiazonic acid
P. islandicum	Cyclochlorotin, luteoskyrin
P. rugulosum	Rugulosin
P. verrucosum	Citrinin, ochratoxin A
Alternaria spp.	
A. infectoria	Alternaria toxins
A. tenuissima	Alternaria toxins
Ustilaginoidea virens	Ustilaginoidins, ustiloxin

In tropical countries where temperature is high throughout the year and frequently paired with high relative humidity (RH), *Aspergillus flavus* is the main fungus in this regard. Recent reports show that the infection of rice grains with *A. flavus* is a chronic problem in some regions and that consumers in these countries can be at risk because of high level exposure to aflatoxins (AFs) (Hussaini, Timothy, Olufunmilayo, Ezekiel, & Godwin, 2007; Katsurayama et al., 2018; Reddy et al., 2009a). In tropical Asia, for example, rice is predominately contaminated with AFs as a result of a pre- and postharvest colonization of the grains with *A. flavus* (Reddy et al., 2009a; Sales & Yoshizawa, 2005; Toteja et al., 2006). In communities where it is consumed as staple food, rice can be considered as the main source of AFs (Park & Kim, 2006; Park, Choi, Hwang, & Kim, 2005). In Japan, the toxicity of moldy rice grains was first reported in 1891 in rice affected by the *Aspergillus* species (Kushiro, 2015).

Aspergillus flavus invasion of rice starts in the field, as it is the case with most cereals, and infection can become worse particularly under poor postharvest practices. In particular, the growth of this fungus on food commodities can be influenced by several intrinsic, extrinsic, implicit, and processing factors (Sinha, 1995); among all, temperature, water activity (a_w), and the air composition are considered as the leading limiting factors for fungal growth and for mycotoxin production in the postharvest stage (Magan, Hope, Cairns, & Aldred, 2003).

Once harvested, rice moisture content plays an important role in fungal development and AF production. Rice with high moisture

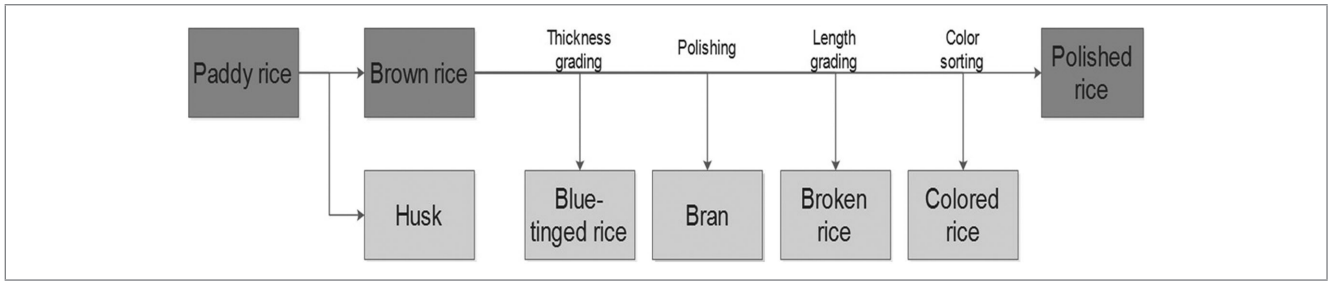

Figure 2–Flow chart of milling process from paddy rice to polished rice (adapted from T. Lee et al., 2011).

content needs to be dried immediately in order to avoid fungal and mycotoxin proliferation. It has been established that *A. flavus* can infect rice grains only when moisture content is higher than 12% (Reddy & Raghavender, 2007).

Growth of different *A. flavus* strains has been tested in paddy rice for temperatures ranging from 10 °C to 43 °C and in a_w ranging between 0.82 and 0.99. None of the tested strains were able to grow neither at the marginal temperatures, regardless of the a_w values, nor at the a_w level of 0.82, regardless of temperature. In contrast, growth on artificial medium was observed at lower a_w level; in particular, Pitt and Miscamble (1995) reported the minimum a_w values for *A. flavus* growth on malt extract agar to be 0.82 at 25 °C, 0.81 at 30 °C, and 0.80 at 37 °C. The estimated optimal growth temperature was around 30 °C (Mousa, Ghazali, Jinap, Ghazali, & Radu, 2011). Differences in growth capacity were also found between polished rice and brown rice with the latter being more prone to *A. flavus* growth and AF production (Sales & Yoshizawa, 2005). The presence of the embryo and the bran layers in the brown rice, which are rich in oils, can give an important advantage to *A. flavus* growth and AF production in comparison with polished rice. Effectively, earlier studies showed that high levels of lipids in the aleurone layer constitute a favorable site for *A. flavus* colonization (Gajapathy & Kalyansundaram, 1986), and the presence of AFs in seeds rich in oil such as cotton, corn, and peanut have validated this hypothesis (Klich, 2007).

The production of AF is markedly a_w-dependent, with a greater level of AF produced at high a_w values (0.95 to 0.98) and lower levels at lower a_w levels (0.92). With inoculation on rice, AF production was observed in the a_w range of 0.86 to 0.99 and the toxin levels were increasing with a_w. Moreover, temperature plays an important role and more AFs are produced at 30 °C compared with a temperature of 20 °C. Water activity shows a greater impact on AF production than temperature, but AF increases as high a_w and high temperature are combined. This has been confirmed also in maize, for which a positive correlation between AF B1 (AFB1) production rate and a_w was found when a_w was higher than 0.95 (Giorni, Bertuzzi, & Battilani, 2016).

Regarding the role of rice varieties, seeds of 11 rice varieties have been studied and none of the varieties were found totally resistant to AF contamination, but differences were noted in the quantities produced (Sales & Yoshizawa, 2005). During an *in vitro* trial, varieties with a high content of starch (713 mg/g) were highly resistant, while, on the contrary, varieties with low starch content (600 mg/g) were the most susceptible to AF accumulation. In particular, a correlation between AFB1, total starch, and amylopectin content in rice varieties has been highlighted (Singh & Sinha, 2013) and was also found in maize studies (Giorni et al., 2015).

Reddy, Raghavender, Salleh, Reddy, and Reddy (2011) tested the potential of AFB1 production by five *A. flavus* strains on cereals (barley, maize, rice, sorghum, and wheat), oilseeds (peanuts and sesame), and pulses (greengram and horsegram), and they concluded that AFB1 accumulation was the highest in rice grains.

Another variable that influences fungal growth and mycotoxin production is CO_2. In particular, a combination of low a_w and high CO_2 can reduce fungal growth rate and increase its lag phase. In another trial, the increment of initial headspace CO_2 from 20% to 80% was associated with a reduction in *A. flavus* growth in rice ranging between 64% and 88%, 59% and 92%, and 55% and 100% at 0.98, 0.95, and 0.92 a_w, respectively. No fungal growth was observed at 80% CO_2 and 0.92 a_w. Regarding AF production, complete inhibition was observed at 0.92 a_w with 80% CO_2. At higher a_w (0.98) and with the same CO_2 level (80%), reduction in AF production ranged between 47% and 99% (Mousa et al., 2016). It has to be underlined that modified atmosphere can be more effective in controlling rice compared with maize colonization by *A. flavus*, because of rice physical structure making it more resistant to the invading fungi and insects compared to maize.

Other mycotoxigenic *Aspergillus* species that have been reported to grow on rice include *Aspergillus ochraceus*, able to produce ochratoxin A (OTA), and *Aspergillus versicolor* which is able to produce sterigmatocystin (STC).

Occurrence of OTA in rice has been reported from different countries with a wide range of occurrence values. Rice samples from Ivory Coast and Turkey were measured for their OTA content and 100% and 30% of samples, respectively, exceeded the European permitted levels (5 μg/kg). However, *A. ochraceus* is generally considered a postharvest problem; because of its ecological needs, when present on grains, it can produce OTA under storage environmental conditions. It has been reported that *A. ochraceus* growth can occur in the range of 8 °C to 37 °C, with the optimum at 24 °C to 31 °C (International Commission on Microbiological Specifications for Foods [ICMSF], 1996). The minimum a_w level for *A. ochraceus* growth has been found at 0.80 to 0.85 on cereal grains (Pardo, Marin, Ramos, & Sanchis, 2006). Ecological studies have revealed that maximum OTA production by *A. ochraceus* occurs at 0.98 a_w at a temperature range of 25 °C to 30 °C. OTA production has been shown to gradually decrease at 20 °C, with even lower values at 10 °C and 5 °C (Ali, Ismail, Bhalli, Mobeen, & Khan, 2013) demonstrating that OTA production can be greatly influenced by temperature (Scheuer & Leistner, 1986). *Aspergillus westerdijkiae*, another important OTA producer, has never been reported in rice. One possible explanation for this is that, only in 2004, *A. westerdijkiae* was separated from *A. ochraceus* and both species are morphologically similar, although the former one is unable to grow at 37 °C and produce white to cream-white sclerotia in contrast with the pink to vinaceous purple sclerotia of *A. ochraceus* (Frisvad, Frank, Houbraken, Kuijpers, & Samson, 2004).

Only a few studies exist on *A. versicolor* in rice. This fungus is generally xerophilic and therefore able to grow at low a_w levels (0.80 a_w), even though its optimal a_w is 0.95. *A. versicolor* can grow in the temperature range of 4 °C to 40 °C with an optimal condition at 30 °C. For STC production, ecological conditions are a little different, with mycotoxin production up to 0.76 a_w in the range of 23 °C to 29 °C (Abramson, Hulasareb, Whitea, Jayasb, & Marquardt, 1999; Atalla, Hassanein, El-Beih, & Youssef, 2003; Rabie, Lubben, & Steyn, 1976). STC can occur in rice and in rice-based products due to fungal infection, especially during the postharvest stage (Biancardi & Dall'Asta, 2015; Uhlig et al., 2013; Veršilovskis, De Saeger, & Mikelsone, 2008).

Fusarium spp. in rice and mycotoxin production

Fusarium spp. are a major cause of reduction in the quality of rice due to environmental conditions during cultivation. High moisture and temperature conditions are favorable for *Fusarium* development in the field. However, apart from being seed-borne pathogens, they may also grow on storage products. They produce different types of symptoms such as seed abortion, seed rot, seed necrosis, and reduction or elimination of seed germination resulting in the development of disease at later stages of growth (Khanzada, Rajput, Shah, Lodhi, & Mehboob, 2002).

Fusarium species are able to produce mycotoxins in rice and these are reported in Table 1.

There are reports of *Fusarium* infection in rice plants worldwide, in particular, due to "Bakanae" disease. Bakanae or foot rot disease is an important emerging disease of rice across the world. Three mating populations of the *Gibberella fujikuroi* species complex have been associated with Bakanae-diseased rice: mating population C (MP-C) (anamorph, *Fusarium fujikuroi*), MP-A (anamorph, *Fusarium verticillioides*), and MP-D (anamorph, *Fusarium proliferatum*). These fungi are taxonomically very similar, and their identification based only on their morphological and cultural features is rather difficult. Thus, more than one species of *Fusarium* may be able to infect rice and cause symptoms of Bakanae disease (Desjardins et al., 2000; Kuhlman, 1982).

The main pathogen responsible for the disease is *F. fujikuroi* (Wulff et al., 2010); however, several studies revealed the occurrence of *F. proliferatum* both in the field (Abbas et al., 1999; Desjardins et al., 2000; Maheshwar & Janardhana, 2010) and postharvest (Park et al., 2005), and the occurrence of *F. andiyazi* in the field (Choi, Hong, Lee, Kim, & Chun, 2018).

Fusaria have been shown to primarily survive in the seeds, but evidence has also shown that it survives in the soil. Currently, seed treatment with fungicides is the most common management practice for Bakanae disease control in India, where basmati and aromatic rice cultivars are regarded as the most susceptible to the disease (Gupta, Solanki, Bashyal, Singh, & Srivastava, 2015). *Fusarium fujikuroi* produces gibberellic acid, fumonisin B1 (FB1), and moniliformin.

Fusarium proliferatum strains have physicopathological effects on rice plants resulting in various symptoms that are able to reduce harvest yield and to accumulate FB1 in rice grains (Abbas et al., 1998; Kushiro, Nagata, Nakagawa, & Nagashima, 2008; Tanaka et al., 2007). *Fusarium proliferatum* infection of rice in ears exhibits different levels of browning. Besides brown discoloration and/or withering in the ears, slight browning can be found at the rachis-branch, lamina joint, leaf sheath, flag leaf, and the tip of the penultimate leaf. This fungus is also able to remain on healthy rice grains under refrigeration for long periods, so that the monitoring of fumonisins in stored rice grains may also be required.

Another *Fusarium* species able to produce mycotoxins in rice is *Fusarium graminearum*, even if rice is not considered a preferential target for this fungus. Generally speaking, grains show the highest susceptibility to *F. graminearum* between the flowering and early stages of the grain (Scotti, Vergoignam, Feron, & Durand, 2001); the fungal contamination may occur in any part of the plant, however, the grains are the most susceptible part (Usha, Patkar, Shetty, Kennedy, & Lacey, 1993). Damage in rice structure is not very frequent (Dors, Primel, Fagundes, Mariot, & Furlong, 2011; Heidtmann-Bemvenuti, Nora, & Badiale-Furlong, 2012) since the external grain portion, formed by lignocellulosic and protein substances, is related to the fungal resistance associated to physical barriers and enzymatic inhibition. The production of trichothecenes, in particular, deoxinivalenol (DON), is a stress consequence promoted by the resistant cultivars (Keller et al., 2013). *Fusarium armeniacum* has also been isolated from rice as another potential producer of trichothecenes, and 24 *F. armeniacum* strains isolated from rice in Korea were able to produce T-2 and HT-2 toxins in potato sucrose agar medium (Hong et al., 2015).

Finally, *Fusarium subglutinans* was also reported in different cereals and resulted to be able to produce the highest amount of different mycotoxins (moniliformin, beauvericin (BEA), and fusaproliferin) on rice (Kostecki et al., 1999).

Penicillium spp. in rice and mycotoxin production

The main *Penicillium* species able to produce mycotoxins in rice are described in Table 1.

Penicillium spp. are frequent contaminants in stored rice. "Yellow rice" is the collective name for rice grain contaminated by *Penicillium* fungi in Japan and refers to grains affected by *Penicillium* species after harvest. This is because no mycotoxin-producing *Penicillium* spp. are found during the growing period in the field (Kushiro, 2015).

Penicillium verrucosum is able to grow on rice during storage. This fungus has the toxicogenic ability to concurrently produce citrinin (CIT) and OTA (Chelkowski, 1985), with two potentially related genes (*pks*CT and *otapks*PN-AS) in CIT and OTA biosynthesis interacting (Geisen, Schimdt-Heydt, Touhami, & Himmelsbach, 2018), and environmental conditions strongly influencing the mutual regulation of CIT and OTA (Schmidt-Heydt, Stoll, Schütz, & Geisen, 2015). Mold development and OTA biosynthesis can be influenced by different factors, such as a_w, temperature, cereal matrix, interaction between mycotoxigenic species, and storage time (Czaban, Wroblewska, Stochmal, & Janda, 2006; Lee & Magan, 2000; Magan et al., 2003; Wawrzyniak, Ryniecki, & Gawrysiak-Witulska, 2013).

Penicillium verrucosum is able to grow in a range of temperatures between 0 °C and 31 °C, with optimum growth at 20 °C (Pardo et al., 2006). Rapid growth of *P. verrucosum* occurs on grain with a moisture content of 27% to 30% (0.97 to 0.99 a_w) over a temperature range between 10 °C and 25 °C (Cairns-Fuller, Aldred, & Magan, 2005). However, accumulation of mycotoxins has also been detected in samples stored between 20 °C and 30 °C. Optimal conditions for OTA and CIT biosynthesis are at 20 °C.

Wawrzyniak and Waskiewicz (2014) tested OTA and CIT production in different cereal crops. Among all cereal substrates (wheat, triticale, rye, barley, maize, and rice), the most favorable matrix for *P. verrucosum* growth, as well as for OTA and CIT biosynthesis, was rice at all tested temperatures. Optimum temperature ranged between 20 °C and 30 °C, and at 10 °C much lower OTA levels were observed, while CIT was only detected in rice. In an *in vitro* trial, at 10 °C, only small amounts of mycotoxins were measured in rice (Häggblom, 1982).

For all rice kernels infected by *P. verrucosum* with an incidence higher than 7%, OTA contamination was shown to be over the European limit of 5 μg/kg (Lund & Frisvad, 2003); however, no linear correlation between OTA content and *P. verrucosum* incidence was observed. In particular, OTA production was concluded to be lower in the case of rapid fungal growth (Häggblom, 1982).

Other fungal diseases in rice and mycotoxin production

The main minor fungal species that are able to produce mycotoxins in rice are reported in Table 1.

Alternaria is a genus of widespread distribution which is capable of producing numerous toxins (AT - *Alternaria* toxins) and these are beyond the scope of this review article. *In vitro* studies showed that *Alternaria tenuissima* and *Alternaria infectoria* strains are able to produce ATs under different environmental conditions, which can be potentially found in real field and storage situations (Zwickel, Kahl, Klaffke, Rychlik, & Mueller, 2016). The production of mycotoxins by *A. tenuissima* strains was demonstrated to be similar in rice and wheat; however, the production by the strain of *A. infectoria* tested was higher (in concentration and number of toxins) in rice.

Another rice fungal disease of growing concern is rice false smut, where spikelets are infected by the fungal pathogen *Ustilaginoidea*

virens (Wang et al., 2017). This fungus produces two families of mycotoxins—ustiloxin and ustilaginoidins—of minor concern.

Occurrence of Multiple Mycotoxins: "Mycotoxin Mixtures"

In rice, the presence and infection with different fungi make unavoidable the co-occurrence of multiple mycotoxins as mixtures of parent compounds and metabolites. A few studies have reported multiple mycotoxin detection in rice and rice products (Table 2). The most commonly reported co-occurring mycotoxins are AFB1 and AFB2, but the co-occurrence of mycotoxins produced by different species has also been documented. Table 2 highlights the latter ones.

Co-occurrence of AFs and OTA has not been clearly reported; however, based on the occurrence of each single mycotoxin, it is clear that some co-occurrence of mycotoxin mixtures exists (for example, Aydin, Aksu, & Gunsen, 2011; Buyukunal, Kahraman, & Ciftcioglu, 2010; Iqbal, Asi, Hanif, Zuber, & Jinap, 2016). *Fusarium* toxins do also co-occur in rice, mainly as DON and zearalenone (ZEN), but evidence for co-occurrence of FB1, T-2 toxin, and HT-2 toxin also exists. Soleimany, Jinap, and Abas (2012) reported their co-occurrence, while the individual co-occurring mycotoxin congeners were not clearly reported.

Majeed et al. (2018) evaluated the presence of 23 mycotoxins in rice, and they detected 9 co-occurring in rice. From 120 samples, the co-occurrence of *Aspergillus* and *Fusarium* mycotoxins was detected in 21% of the samples.

In processed rice products, the co-occurrence has not been well documented, and most of the information is related to the co-occurrence of mycotoxins in red yeast rice (RYR) (Samsudin & Abdullah, 2013). Nevertheless, this is a rather susceptible product for AF occurrence and high levels have been detected (Katsurayama et al., 2018).

Cropping System for Mycotoxin Prevention

Globally, rice yields can range from less than 1 ton per hectare (t/ha) from poor rain-fed cropping systems to as much as 10 t/ha from irrigated and intensive temperate-region rice cultivation (International Rice Research Institute [IRRI], 2013). Irrigated lowland cultivation is the most important rice cropping system for food security, and it occupies 79 million hectares worldwide covering around 75% of the total world rice production area (IRRI, 2013).

Rice-based cropping systems in irrigated or favorable environments in Asia and Africa have been intensified to also grow wheat and maize. This has been necessary to optimize use of resources, since the diversification of rice systems in rotation with other cereals (like wheat or maize) or with high-value crops (like potato, legume, or fodder crops) can increase land productivity and minimize unpredictable risks such as the build-up of pests and diseases common in a rice monoculture (IRRI, 2013). However, in some countries where the land dedicated to rice production is restricted (like Italy or Spain), monoculture is usually the norm.

Agronomic and crop management strategies aiming to control Fusarium head blight (FHB) do consider foliar fungicide applications and tillage practices; however, these are generally not very effective (Martin, MacLeod, & Caldwell, 1991; McMullen, Jones, & Gallenberg, 1997; Parry, Jenkinson, & McLeod, 1995).

Cereal rotation and lodged fields seem to reduce *Fusarium* infestations and mycotoxin presence (Bernhoft, Torp, Clasen, Løes, & Kristoffersen, 2012), while occurrence of mycotoxins seems to increase when using organic agricultural methods (Ok, Choi, Chang, Chung, & Chun, 2011; Rubert, Soriano, Mañes, & Soler, 2013; Serrano, Font, Mañes, & Ferrer, 2013). However, evidence for the impact of cropping systems on mycotoxin occurrence and, consequently, on health risk factors is still controversial (Oliveira, Zannini, & Arendt, 2014).

In this context, the use of resistant rice varieties may be an effective way to reduce mycotoxin contamination; however, only a few rice blast-resistant varieties have been developed until now (Deng et al., 2017; Khush & Jena, 2009).

Technologies applied in precision agriculture can play an important role in field management, and in pest and disease control. In particular, the use of satellite can be useful to better distribute iron to crops, and improve seed and fertilizer distributions. Sprinklers that are able to open or close automatically, avoiding overpassing, can reduce pesticide use, and support an economic return and safer productions (Miserocchi, 2018).

Possible Reduction of Mycotoxigenic Fungi in the Field

Chemical control of mycotoxigenic fungi

Only a few studies have been conducted with regard to control strategies for mycotoxigenic fungi in rice fields. In general, chemicals have been shown to be efficient in crop protection (Lamberth, 2009), but they may have many negative effects. While acidifying the soil and, consequently, decreasing the occurrence of beneficial organism populations, they may potentially interfere with the plant's growth (Oliveira et al., 2014). Moreover, appearance of fungicide-resistant pathogenic strains suggests the need to find alternative methods to chemicals for controlling plant diseases in the field (Katagiri, Uesugi, & Umehara, 1980; Suzuki, Yamaguchi, Koba, Nakajima, & Arai, 2010). In agricultural systems with high levels of sustainability, there is currently increasing pressure to reduce the use of insecticides, fungicides, and herbicides (Chandler, 2008), particularly because these can even have adverse effects on human health and the environment (World Health Organization [WHO], 1990).

For rice, some commercial fungicides, such as for seedling treatments, have been evaluated for their efficacy against Bakanae disease caused by *Fusarium* species under natural field infection and after artificial inoculation (Bagga & Sharma, 2006). Of the five fungicide formulations that have been assessed under natural field infection, those based on carbendazim, benzimidazole, and propiconazole were able to significantly reduce Bakanae incidence and improve grain yield (Bagga & Sharma, 2006). Efficacy of carbendazim and benzimidazole on Bakanae control has also been found in previous studies (Ou, 1985; Titone, Polenghi, Tamborini, & Garibaldi, 2004).

Treatment with propiconazole was the most effective in reducing the occurrence of the disease, but it caused reductions in plant height, tillering, and in grain yield. The other two fungicides applied (one containing mercury and the other containing thiophanate-methyl) were poorly effective on reducing the spread of the disease. Considering the artificial inoculation of seeds, all the five fungicides significantly reduced foot rot, and improved plant height and grain yields (Bagga & Sharma, 2006).

FHB caused by *F. graminearum* is generally managed with triazole applications that are able to reduce both disease and DON occurrence (Jordahl, Meyer, & McMullen, 2010; Ransom, Pederson, & Halley, 2010). The application of fungicide is most effective if sprayed prior to infection; however, under environmental conditions that are particularly favorable for disease

Table 2–Occurrence of mycotoxins in rice and derived products.

Mycotoxin	Commodity	Observation	Reference
AF; OTA	Rice and products	- High co-occurrence of AFB1 and AFB2 - Not clear if OTA co-occur with AFs, but strong evidence of co-occurrence in brown rice	(Iqbal et al., 2016)
	Rice	- Co-occurrence of AFs and OTA	(Aydin et al., 2011)
	Rice	- Strong evidence of co-occurrence	(Buyukunal et al., 2010)
AF; FB1; DON	Rice	- Co-occurrence of DON and FB1 - Co-occurrence of AFs and FB1	(Ortiz, Van Camp, Mestdagh, Donoso, & De Meulenaer, 2013)
AF; OTA; ZEN	Rice	- Not clear which mycotoxins co-occur	(Rahmani, Jinap, & Soleimany, 2010)
AFB1; OTA; FB1	Rice	- Co-occurrence clearly stated	(Bansal et al., 2011)
AF; OTA; DON; ZEN	Rice	- Strong evidence but not clear which mycotoxins co-occur	(Abdulkadar, Al-Ali, Al-Kildi, & Al-Jedah, 2004)
AFB1; OTA; DON; ZEN	Rice	- Co-occurrence of DON and OTA in polished rice - Co-occurrence of DON and ZEN in parboiled rice and white bran - Co-occurrence of OTA and ZEN in natural rice in husk - Co-occurrence of AF, OTA, DON, and ZEN in parboiled bran	(Dors et al., 2013)
AFB1; OTA; DON; ZEN; CTV	Rice and byproducts	- Co-occurrence of AF and ZEN in 17.0% of samples - Co-occurrence of AF and OTA in 24.2% of samples - Co-occurrence of AF and CTV in 6.2% of samples - Co-occurrence of OTA and CTV in 4.6% of samples - Co-occurrence of ZON and CTV in 3.1% of samples	(Almeida et al., 2012)
AF; OTA; ZEN; DON; FB; T2; HT2	Rice	- Not clear which mycotoxins co-occur	(Soleimany et al., 2012)
AF; OTA; FB1; DON; ZEN	Rice	- Co-occurrence of the mycotoxins	(Egbuta, Wanza, & Dutton, 2015)
AF; OTA; DON; NIV; DAS; FB; ZEN; HT-2	Rice	- 78% of samples with at least one mycotoxin - 57% of samples with co-occurring mycotoxins - 40% of samples with AFB1 and AFB2 - 21% of samples with co-occurring *Aspergillus* and *Fusarium* toxins	(Majeed et al., 2018)
AF; OTA; CIT	Red yeast rice	- Co-occurrence of the mycotoxins	(Samsudin & Abdullah, 2013)

development, infection and DON contamination cannot be avoided (Oliveira et al., 2014).

The effect of propionic acid and a commercial antifungal agent containing propionic acid—Monoprop—on the occurrence of AFB1 levels showed reductions by 100% (by applying 0.05 μg/kg or 0.10 μg/kg) and 50% (for 2 μg/kg) for each treatment, respectively (Bedi & Agarwal, 2014). However, it is noted that this study focused on feed, and the applicability of the results in the field still needs to be demonstrated.

In an in-field study, Dors et al. (2013) studied the application of the fungicide tebuconazole (0.75 L/ha) in irrigated rice. These authors found a positive trend between mycotoxin content (AF, OTA, DON, ZEN) and fields with fungicide application, suggesting that the fungicide acted as a stress factor, inducing mycotoxin productions and, consequently, their accumulations. This observation led to the conclusion that the choice of the fungicide should not rely just on plant productivity and plant disease, but also on the mitigation of mycotoxin production.

Biocontrol of mycotoxigenic fungi

With regard to possible alternative methods to limit fungi development and mycotoxin production, different microorganisms have been tested. These microorganisms may control plant diseases through one or more mechanisms, including competition with pathogens for space and nutrients, the production of antimicrobial compounds, the induction of host resistance to the disease, or direct antagonism to the pathogens (Compant, Duffy, Nowak, Clement, & Barka, 2005). In this context, even microbial interactions in the rhizosphere have been shown to contribute to plant crop biocontrol (Barea, Pozo, Azcón, & Azcón-Aguilar, 2005; Whipps, 2001).

Among the microbial populations tested, lactic acid bacteria (LAB) seem to have a great potential as an agent to control fungal diseases. In particular, spraying diluted solutions of LAB onto the plant and soil have been hypothesized to support healthy plant growth (Oliveira et al., 2014). Biocontrol agents have been tested as control agents for cereal diseases caused by *Fusarium* species (Khan & Doohan, 2009; Khan, Fischer, Egan, & Doohan, 2006) and the results showed efficacy in reducing *Fusarium* contamination (Lowe & Arendt, 2004; Reinikainen, Peltola, Lampinen, Haikara, & Olkku, 1999).

Among the tested microorganisms (Table 3), *Streptomyces corchorusii* strain UCR3-16 has been studied for its ability to produce cell wall degrading enzymes and diffusible and volatile compounds. With this strain, inhibition of different rice pathogens has been achieved, among them of one strain of *Fusarium oxysporum* (MTCC 287) (Tamreihao et al., 2016).

While studying different *Trichoderma* strains *in vitro*, *Trichoderma gamsii* 6085 was efficient in reducing *Fusarium culmorum* and *F. graminearum* in rice kernels, with a simultaneous inhibition of DON production with no negative impact of the pathogen's presence on the survival of the biocontrol agent (Matarese, Sarrocco, Gruber, Seidl-Seiboth, & Vannacci, 2012).

Burkholderia gladioli M3, a bacterium that is often present in rice, co-occurring with *A. flavus*, has also been studied for its antagonistic effects on *A. flavus* growth and spore production, and results have demonstrated inhibition by using a cell-free culture filtrate and by a metabolite of *B. gladioli* M3 (bongkrekic acid) (Quan-Hong et al., 2015). Simultaneously, no effect of *A. flavus* was detected on the antagonistic strain.

Postharvest Strategies

Storage

Storage conditions play an important role in mycotoxin control, since they will influence overall fungal development. In general, high humidity and temperature can favor fungal growth and

Table 3–Biocontrol microorganisms and their metabolites with inhibitory potential on mycotoxin-producing fungi.

Microorganism and metabolite	Target fungal species	Type of assay	Reference
***Bacillus subtilis, Pseudomonas fluorescens, Rhodococcus erytropolis,* and *Trichoderma virens* culture filtrates**	*A. flavus*	*In vitro* (naturally infected rice seeds)	(Reddy et al., 2009b)
***Burkholderia gladioli* M3 cells and culture filtrates**	*A. flavus*	*In vitro*	(Quan-Hong et al., 2015)
***Streptomyces corchorusii* UCR3-16 cells**	*F. oxysporum* (MTCC 287)	*In vitro*	(Tamreihao et al., 2016)
***Trichoderma gamsii* 6085 cells**	*F. culmorum* and *F. graminearum*	*In vitro* (rice kernels)	(Matarese et al., 2012)

promote mycotoxin production. Storage under controlled conditions, such as packaging practices, temperature control, ventilation efficiency, and proper air humidity, will reduce fungal development and mycotoxin accumulations (Hina, Shahid, & Ali, 2014).

Just by controlling temperature and moisture of grains, a safer storage can easily be promoted. The assessment of moisture content of grains and storage temperature of long-grain rough rice has indicated that safe storage could be achieved for 6 weeks at temperatures and moistures below 27 °C and 17%, respectively (Atungulu, Thote, & Wilson, 2016). For *A. flavus* in rice, population growth has been demonstrated to remain constant for 120 days of storage at 21 °C and 85% RH. At 21 °C and 97% RH and at 30 °C and 85% RH, fungal growth and AFs' production were mostly influenced by high humidity, and most significantly in brown rice, compared with rough or white rice (Choi et al., 2015). The same experiment for *F. graminearum* showed that at 85% RH (both at 21 °C and 30 °C) neither fungal development nor DON production were detected. In contrast, with 97% RH, even at 21 °C, fungal populations and mycotoxin production were detected in the three types of rice tested (Choi et al., 2015).

Application of a range of techniques to inhibit *A. flavus* growth in stored rice grains showed that after 15 months, approximately 67%, 63%, 37%, and 6% of inhibition were achieved through the application of potassium sorbate, turmeric (*Curcuma longa*), clove (*Syzygium aromaticum*), and microwave heat, respectively (Anuja & Gupta, 2010).

In addition, storage of rice grains in polylined jute bags was found to reduce fungal incidence more efficiently when compared with jute bags (Anuja & Gupta, 2010). The application of ultra-hermetic airtight containers seems to be an effective way to store grains, confirmed to overcome the risk of AF and fumonisin contaminations, and also promising for the other mycotoxins since these inhibit fungal growth while taking advantage of the accumulation of CO_2 and the reduction of O_2 resulting from respiration (Uegaki, Kobayashi, Inoue, Tohno, & Tsukiboshi, 2013; Villers, 2014).

Modified atmospheres with varying CO_2 concentrations can protect rice from fungal infestation. Concentrations of 60% reduced the incidence of storage fungi (for example, *Cladosporium* spp. and *Alternaria alternata*), but 80% was required to control *A. flavus* (Anuja, Sinha, Atwal, & Gupta, 2014). With reduced O_2 concentration below 1%, at all a_w levels studied, the growth parameters, as estimated by Baranyi function, and the AF content were affected by the increment in CO_2, where growth rates and AF were negatively correlated with CO_2, while the lag phase duration was positively correlated with CO_2 (Mousa et al., 2016).

Sugihara et al. (2016) studied production of the AFs in rice during storage under natural climate conditions in Japan. After artificial inoculation with *A. flavus*, these authors found that AF production took place during storage, with higher levels detected in white rice than in rough rice. The storage of rice as rough rice could be a useful means for the prevention of AF contamination.

Naturally occurring compounds to prevent mycotoxin production

Among a vast array of naturally occurring compounds with antifungal properties, botanical essential oils have received an increasing interest over the past decade (Table 4). These complex botanical compounds have been tested to prevent growth of mycotoxigenic strains and to prevent the production of mycotoxins. However, trials have only been performed on a small scale, mostly under *in vitro* conditions.

Basil (*Ocimum basilicum*) essential oil treatment at 2% (v/wt) in inoculated rice resulted in log reductions of 1.18 and 1.12 for the growth of *A. niger* and *P. chrysogenum* after 14 days, respectively. When applied in association with gamma-radiation treatments, *O. basilicum* oil increased the efficacy of irradiation, with reductions of 4.6 and 5.0 log after the combined treatment, for each species, respectively (Hossain et al., 2014).

Reddy, Reddy, and Muralidharan (2009b) tested 9 plant extracts in the treatment of naturally infected rice seeds, and flower buds of *S. aromaticum* completely inhibited *A. flavus* growth at 5 g of flowers per kg rice seeds. At a lower concentration of 3 g/kg, an inhibition of 55% and 98% of *A. flavus* growth and AFB1 production was observed, respectively. A later study confirmed the activity of an oil from *S. aromaticum* on *A. parasiticus* growth (Suganthi, Manpal, David, & Mech, 2013).

Waste fruit peels have also been tested for their inhibitory potential against an *A. flavus* strain. The whole fruit peels, as well as their ethanolic and methanolic extracts, were active against fungal growth. Mycotoxin production by *A. flavus* in inoculated basmati rice mixed with powdered fruit peels was completely inhibited with pomegranate peels and lemon peels after 4 and 3 months, respectively (at 18% moisture and 25 °C) (Naseer, Sultana, Khan, Naseer, & Nigam, 2014).

Processing

Rice processing includes milling to different degrees of coarseness to obtain a range of final products. Starting from the rough rice, hulls can be removed to get brown rice, and then milling allows the removal of the outer bran to obtain white rice (Trucksess, Abbas, Weaver, & Shier, 2011). This process is relevant when dealing with contamination risk, since the distribution of mycotoxins in these fractions is different. Studying AFB1 and AFB2 starting from rice seeds (paddy rice), the greater fractions of mycotoxins have been found to be in brown rice and bran, whereas the lowest contamination was found in white rice, indicating that the most significant step to overcome AFs is the removal of bran to obtain white rice (Trucksess et al., 2011). Also for DON, nivalenol (NIV), and ZEN, lower mycotoxin loads were found in polished rice compared to brown rice, and two byproducts (blue-tinged rice and discolored rice) were heavily contaminated with the mycotoxins (Lee et al., 2011).

During the rice milling process, Dors et al. (2013) studied the distribution of mycotoxins between husk, bran, and starchy

Table 4–Plants with naturally occurring compounds with inhibitory potential on mycotoxin-producing fungi.

Plant	Target fungal species	Type of assay	Reference
Acacia nilotica, *Caesalpinia coriaria*, *Decalepis hamiltonii*, *Emblica officinalis*, *Lawsonia inermis*, and *Mimosops elengi* methanolic extract	*A. alternata*, *A. flavus*, *Curvularia lunata*, *Drechslera oryzae*, *Drechslera halodes*, *F. moniliforme*, *Pyricularia oryzae*, and *Trichoconis padwickii*	*In vitro* (CzA)	(Mohana, Prasad, Vijaykumar, & Raveesha, 2011)
Allium sativum, *Curcuma longa*, *Ocimum sanctum*, and *Syzigium aromaticum* extract	*A. flavus*	*In vitro* (naturally infected rice seeds)	(Reddy et al., 2009b)
Alpinia conchigera and *Alpinia galanga* methanolic extract	*A. flavus*	*In vitro* (CYA)	(Yazdani, Abidin, Tan, Kamaruzaman, & Jaganath, 2012)
Andrographis paniculata, *Cymbopogon citratus*, *Eurycoma longifolia*, *Kaempferia galanga*, and *Orthosiphon aristatus* aqueous extract	*P. citrinum*	*In vitro* (PDB)	(Reddy, Nurdijati, & Salleh, 2010)
Azadirachta indica, *Citrus limonum*, *Cyanodon dactylon*, and *Ocimum sanctum* fresh leaves	*A. parasiticus*	*In vitro* (PDA)	(Suganthi et al., 2013)
Azadirachta indica phenolic extract from seeds; rice bran γ-oryzanol and phenolic extract	*F. graminearum*	*In vitro* (PDA)	(Heidtmann-Bemvenuti, Tralamazza, Ferreira, Corrêa, & Badiale-Furlong, 2016)
Brassicacea allylisothiocyanate and *Syzygium aromaticum* essential oil	*F. graminearum* and *A. westerdijkiae*	*In vitro* (PDA and CYA)	(Cardiet, Fuzeau, Barreau, & Fleurat-Lessard, 2012)
Capsicum frutescens and *Zingiber officinale* pulps	*A. parasiticus*	*In vitro* (PDA)	(Suganthi et al., 2013)
Curcuma longa, *Myristica fragrans*, *Piper nigrum*, and *Terminalia chebula* plant materials	*A. parasiticus*	*In vitro* (PDA)	(Suganthi et al., 2013)
Cymbopogan citratus, *Cymbopogan martini*, and *Pelargonium graveolens* essential oil	*F. moniliforme* and *Helminthosporium oryzae*	*In vitro* (PDA)	(Muthukumar, Sangeetha, & Naveenkumar, 2016)
Cymbopogan citratus, Cymbopogan martini, and *Syzygium aromaticum* essential oil	*A. parasiticus*	*In vitro* (PDA)	(Suganthi et al., 2013)
Laurus nobilis and *Syzygium aromaticum* essential oil	*F. culmorum*, *P. islandicum*, *A. candidus* and *A. niger*	*In vitro* (PDA)	(Magro et al., 2010)
Lemon-peels and pomegranate-peels	*A. flavus*	*In vitro* (PDA) and in inoculated rice samples	(Naseer et al., 2014)
Michelia alba essential oil	*A. flavus*	*In vitro* (MEA) and inoculated brown rice	(Sumethee, Narumol, & Nirundorn, 2017)
Ocimum basilicum essential oil	*A. niger* and *P. chrysogenum*	*In vitro* (PDA) and inoculated rice samples	(Hossain et al., 2014)
Syzygium aromaticum extract	*P. citrinum*	*In vitro* (YES) and in white polished rice	(Aiko & Alka, 2013)

Note: CzA = Czapek-Dox Agar; CyA = Czapek Yeast Agar; PDA/PDB = Potato Dextrose Agar/Broth; MEA = Malt Extract Agar; YES = Yeast Extract Sucrose Agar.

endosperm. AF and DON were distributed among all fractions (even after the parboiling process), while OTA and ZEN were mainly found in bran and husk. These data suggest that the parboiling process may contribute to the migration of some mycotoxins into the rich starchy endosperm (as observed before by the same authors (Dors, Pinto, & Badiale-Furlong, 2009)).

Cooking of rice reduced the levels of AFs. According to Majeed et al. (2018), initial washing of rice reduced about 15% AFs levels, and subsequent cooking reduced AFB1 (41% to 51%) and AFB2 (47% to 63%), depending on the cooking process and with significantly higher reduction when excess water is drained after cooking.

FB1 levels in rice have been assessed after thermal treatments, consisting of cooking, autoclaving, and dry heat, and results showed that conventional cooking resulted in a reduction of up to 80%, autoclaving for 22.5 min with soaking for 5 hr reduced levels by up to 74% and heat treatment by a maximum of 82.8% (Becker-Algeri, Heidtmann-Bemvenuti, Hackbart, & Badiale-Furlong, 2013). The experiments were designed for three types of rice, and the results demonstrated that conventional cooking was more effective in reducing FB1 levels in parboiled rice. These can be associated to the gelatinization of starch occurring during the parboilization process, which enables a more efficient contact with water and a consequent reduction of mycotoxin content in rice (Becker-Algeri et al., 2013).

Kim, Scott, Lau, and Lewis (2002) showed that FB1 and FB2 were unstable when added to Thai white rice flour and that they disappeared completely after 10 hr at room temperature. The rationale for this is yet to be evidenced; however, a possible explanation can be related to the interaction of FB with starch, preventing accuracy of the analytical method.

The stability of BEA during bread preparation has been studied using different flour types. Bread prepared from rice flour showed nearly a 50% reduction at 160 °C (20 min of incubation), and about 60% and 80%, at 180 °C and 200 °C, respectively; with lower degradations in food systems when compared to the complete degradation achieved in a model system (Meca, Ritieni, & Manes, 2012).

Originating from China, the *Monascus purpureus* fermentation of cooked rice kernels to obtain RYR, or red mold rice, is commonly used for its preservative, coloring, and medicinal properties. Mainly for its health benefits by lowering cholesterol levels, there is an increasing interest for RYR around the world (Kalaivani, Sabitha, Kalaiselvan, & Rajasekaran, 2010). In RYR, the levels of monacolin K, which is formed during the fermentation process and inhibits HMG-CoA reductase (involved in cholesterol synthesis), are crucial to achieve the desirable functional properties, but since fungal activity is also associated with the production of CIT, there is a demand for the optimization of the process (Kalaivani et al., 2010; Kiebooms, Huybrechts, Thiry, Tangni, &

Callebaut, 2016; Liao, Chen, Lin, Chiueh, & Shih, 2014). The selection of *Monascus* strains and the application of mutagenesis techniques to increase the monacolin K/CIT ratio produced during fermentation are being explored (Kanpiengjai, Mahanwan, Pengnoi, Lumyong, & Khanongnuch, 2018; Tsukahara, Shinzato, Tamaki, Namihira, & Matsui, 2009). Kanpiengjai et al. (2018) obtained a mutant strain capable of increasing the M/C ratio of production in RYR obtained from glutinous rice, from 1450 to 1790. Besides, the optimization of the solid-state fermentation variables allowed a further increase to 3900 (adopting an incubation time of 38 days and a medium's moisture content of 75%). In another study, the methods of dry heating, wet heating, hydrogen peroxide detoxification and extraction with alkaline solution, ethanol, and phosphate-ethanol were applied under different conditions and results were compared (Lee, Chen, Wang, & Pan, 2007). Overall, the highest M/C ratios (calculated from the percentage of compounds' retention during the treatments) were obtained using the extractions with alkaline solution (5 mL, 0.25 M sodium carbonate, 60 min, 30 °C), ethanol (10 mL, 30% ethanol, 60 min, 65 °C), and phosphate-ethanol (45% ethanol, 1.5 % phosphate, 70 min, 65 °C), with the latter increasing the M/C ratio 9.46 times (Lee et al., 2007). Additionally, the application of physical or chemical methods to detoxify the product with a minimum impact on monacolin K levels has shown promising results (Kanpiengjai et al., 2018; C. Lee et al., 2007; Nannoni, Ali, & Di Pierro, 2015).

Experimental Strategies for Decontamination

The mycobiota of commodities include different field fungi, but, after harvest, *Aspergillus* and *Penicillium* are the most prevalent mycotoxigenic ones (Mannaa, Kim, & Kim, 2016). Although different strategies have been presented before to avoid their occurrence, techniques to reduce mycotoxin levels constitute an alternative in situations where their occurrence cannot be completely avoided. Bedi and Agarwal (2014) showed AFB1 reductions by up to 17% after directly exposing basmati rice to sunlight. Also, reductions on AFB1 levels by 40% to 46% were detected when extracts of artificially contaminated rice were exposed to 1% activated charcoal, reaching up to 95% of toxin decrease with 4% of activated charcoal (Bedi & Agarwal, 2014).

Pulsed-light treatments have degradation potential against AFBs. After application at 0.52 J/cm^2/pulse, reductions of 75% and 39.2%, for rough rice after 80 seconds, and of 90.3% and 86.7%, for rice bran after 15 s, were obtained for AFB1 and AFB2, respectively (Wang et al., 2016). AFB1 degradation can also be achieved by rice treatments with γ-irradiation, which were shown to provide reductions of more than 95% (at 6 kGy) from the initial levels, and, except for a decrease in oleic acid and an increase in leucine, amino acid and fatty acid contents were not significantly affected by this treatment (Ahsan, Hussain, Naqvi, & Asi, 2013). In case of irradiation treatments of inoculated rice samples, reductions of 2.18 log and 2.41 log for *A. niger* and *P. chrysogenum* incidences were obtained at 2 kGy, respectively. The efficacy of irradiation increased in the presence of *O. basilicum* essential oil at 2% (v/wt), with reductions of 4.6 log and 5.0 log (Hossain et al., 2014).

Ozone fumigation is also known to cause degradation of mycotoxins; however, the method of application influences differently the physical and biochemical characteristics, as well as reduction rates. A study with AFB1 in paddy rice showed that ozone fumigation using a dry method caused a degradation of about 70%, while aqueous and wet methods reduced mycotoxin by about 87% and 94%, respectively. Also, wet methods slightly increased water content and reduced rice germination rate (Wang, Liu, Lin, & Cao, 2010).

Benzyl isothiocyanate and phenyl isothiocyanate were also studied for their decontamination properties, showing reduction of BEA levels, both in a model solution and in cereal matrixes. BEA reductions of 92.5% and around 90% in rice kernels, and of 65.5% and 70.3% in rice flour were obtained for benzyl isothiocyanate and phenyl isothiocyanate, respectively (Luciano, Meca, Manyes, & Manes, 2014).

The application of bacteria to degrade CIT present in RYR due to *Monascus* spp. was also studied. It was found that among five isolates that could degrade CIT without affecting the desirable monacolin K levels, *Enterococcus cloacae* and *Rhizobium borbori* were able to decrease CIT from 5 ppm to 1.83 ppm and 2.82 ppm, after 120 hours at 30 °C, respectively (Kanpiengjai, Mahawan, Lumyong, & Khanongnuch, 2016). Interestingly, also some *Aspergillus* strains seem to be able to detoxify mycotoxins, in this case through enzyme activity, as it was shown in a study that attested *Aspergillus welwitschiae* ability to transform fumonisins in nonaminated forms (FPy and FLa), which are much less toxic, addressing possible new approaches in decontamination processes (Burgess, Renaud, McDowell, & Sumarah, 2016).

Conclusion

Fungal contamination in rice is a factor of major importance when assessing rice safety for human and animal consumption. Because of the importance of rice as a food commodity, further highlighted by high intakes in many regions of the world, assessing and lowering the risks of mycotoxin occurrence resulting from fungal infection in rice is crucial. Indeed, different fungi present in rice can be a threat at many levels of the rice food chain, starting from the field to the final stages of processing. In addition, the different ecological needs of the different fungal genera make them a constant hazard and the occurrence of multiple mycotoxins as parent compounds or metabolites measured in rice and processed products further highlight this matter. Appropriate strategies to mitigate mycotoxin contamination in rice include the control of the agronomic and postharvest practices, taking into account that processing can reduce mycotoxin contents, but not completely overcome their occurrence in the final product.

Predictive modeling was applied in other crops, mainly wheat and maize, as valuable support to crop management in a whole food chain view (Battilani, 2016; Battilani & Camardo Leggieri, 2015), but never considered for rice and mycotoxin-producing fungi. Modeling should be included in the next future as a hot topic for research, also on account of the climate change we are facing at a global level. In fact, there can be expected a strong impact on plant biogeography, and also shifts in fungal populations and mycotoxin patterns, as has already been confirmed for AFs (Battilani et al., 2016; Van der Fels-Klerx, Liu, & Battilani, 2016).

Acknowledgments

This review was prepared as part of MYCHIF EFSA project (GP/EFSA/AFSCO/2016/01). Roberta Palumbo carried out this work within the PhD school Agrisystem of Univ. Cattolica del Sacro Cuore (Italy). This study was also supported by the Portuguese Foundation for Science and Technology (FCT) under the scope of the strategic funding of UID/BIO/04469 unit and COMPETE 2020 (POCI-01-0145-FEDER-006684) and BioTecNorte operation (NORTE-01-0145-FEDER-000004) funded by the European Regional Development Fund under the scope of Norte2020 - Programa Operacional Regional do Norte.

Authors' Contributions

A. Gonçalves: literature review; paper writing; P. Giorni: literature review; paper writing and revision; A. Gkrillas: literature review; J. L. Dorne: design of the objectives of the project, paper review and revision; R. Palumbo: literature review; C. Dall'Asta: paper revision; N. Lima: paper revision; P. Battilani: conception and design, paper revision. A. Venâncio: paper preparation, paper coordination, revision, and final approval. All authors provided critical feedback and helped shape the manuscript.

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