Does the Host Contribute to Modulation of Mycotoxin Production by Fruit Pathogens?

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Abstract: Storage of freshly harvested fruit is a key factor in modulating their supply for several months after harvest; however, their quality can be reduced by pathogen attack. Fruit pathogens may infect their host through damaged surfaces, such as mechanical injuries occurring during growing, harvesting, and packing, leading to increased colonization as the fruit ripens. Of particular concern are fungal pathogens that not only macerate the host tissue but also secrete significant amounts of mycotoxins. Many studies have described the importance of physiological factors, including stage of fruit development, biochemical factors (ripening, C and N content), and environmental factors (humidity, temperature, water deficit) on the occurrence of mycotoxins. However, those factors usually show a correlative effect on fungal growth and mycotoxin accumulation. Recent reports have suggested that host factors can induce fungal metabolism, leading to the synthesis and accumulation of mycotoxins. This review describes the new vision of host-factor impact on the regulation of mycotoxin biosynthetic gene clusters underlying the complex regulation of mycotoxin accumulation in ripening fruit.

Keywords: mycotoxin; fruit pathogen; food safety; fungal pathogenicity; fruit ripening

1. Introduction

Fruit spoilage by fungi causes not only substantial economic losses but also health risks upon mycotoxin consumption. Mycotoxins are toxic low-molecular-weight secondary metabolites produced mainly by filamentous fungi belonging to the genera Aspergillus, Penicillium, Alternaria, and Fusarium. Mycotoxins, which are secreted with only minor effects on fungal growth onto various foods and feeds, can cause diseases collectively known as mycotoxicoses in humans and animals upon ingestion [1]. The consumed metabolites can affect single or multiple target organs, leading to cytogenic, mutagenic, carcinogenic, teratogenic, or immunosuppressive effects [2]. Hence, in many countries, mycotoxin levels in foods are regulated, and the U.S. Food and Drug Administration limits mycotoxin levels in fruit products [3,4]. This review analyzes the recently discovered factors modulating the occurrence and regulation of mycotoxins in postharvest fruit and provides examples of how the fruit host might modulate the induction of mycotoxin production.

Fruits may become infected with mycotoxigenic fungi through damaged surfaces—mechanical injuries, insect wounds, cuts, and splits—that occur during growing, harvesting, packing, transporting, postharvest storage, and marketing. Toxin production is dependent on a variety of complex interactions.
between internal and environmental factors, including the geographical location where the fruit is grown and harvested, the humidity, temperature, pathogen load on the fruit, fungal strain, fruit type and cultivar, ripening, and fruit physiological properties [5,6]. Fruit maturity at harvest is believed to be an important factor in susceptibility to infection by pathogenic fungi during postharvest storage, due to high sugar content, water activity \((a_w)\), changes in pH, decreased firmness, and weakening defense systems [7–9].

There are large numbers of known mycotoxins but only a few are commonly found in fruit: aflatoxins in dried figs \((Aspergillus flavus)\), ochratoxin A in grapes, peaches, cherries, strawberries, apples, and dried figs \((Penicillium and Aspergillus\) species), patulin in deciduous fruits \((Penicillium and Aspergillus\) species), alternariol in tomatoes, apples, tangerines, and mandarins \((Alternaria\) species), citrinin in deciduous fruits \((Penicillium\) species) and trichothecenes in sweet pepper \((Fusarium and Trichothecium\) species) [6,10–12]. However, few studies have examined the importance of fruit metabolic changes during ripening in mycotoxin production. Since the infection by mycotoxigenic fungi in fruit occurs in the field, during harvesting, postharvest, and during storage, the physiological changes inducing ripening occurring in the host after infection and their contribution to mycotoxin accumulation are likely of great importance.

2. Mycotoxins Commonly Found in Fruit

2.1. Patulin

Patulin is a low-molecular-weight \(\alpha, \beta\)-unsaturated \(\gamma\)-lactone found in fruit. This toxin causes severe toxicity due to neurotoxic, nephrotoxic, mutagenic, teratogenic, or hepatotoxic effects, which can result in nausea, vomiting, kidney damage, and gastrointestinal disorders [13–15]. \(Penicillium expansum\), an airborne fungus, is the major species contributing to patulin accumulation in apple products [16]. Several other fungi can produce patulin, including \(Aspergillus clavatus\), \(A. giganteus\), \(A. terreus\), \(Byssochlamys fulva\), \(B. nivea\), and several \(Penicillium\) species [17–20]. Patulin can be found in several types of mold-colonized fruit, including pears, plums, berries, and tomatoes, but the major source of patulin contamination is apples and its juice products [21–23].

Factors Affecting Patulin Production

Various factors are known to affect patulin production in apple, including cultivar type, geographical location, climate, mechanical injury, storage conditions, and pre- and post-harvest conditions.

**Fruit development conditions**—As it matures, the fruit undergoes physiological changes, such as increases in pH and total soluble solids (TSS), decreases in firmness and acidity, and weakening of the defense system, which can increase its susceptibility to pathogen and patulin production [8,9]. Moreover, patulin accumulation in apples is affected by environmental conditions, fungicide residues, pesticide treatments, microbial load, harvest method, and postharvest treatments [24]. Foliar spray of chemicals on apple trees during fruit development results in significant reductions in \(P. expansum\) infection and consequently, in mycotoxin accumulation [25–27].

**Fruit cultivar**—Several reports concluded that fruit cultivars differ in their susceptibility to pathogen attack and patulin production in apples [28–33]. Fruit cultivars differ in their physical and chemical properties, such as skin thickness, pH, TSS, acidity, firmness, and defense systems, which may affect patulin production [29,32,34]. A study of four apple varieties showed that patulin accumulation is significantly higher in ‘Red Delicious’ and ‘Golden Delicious’ than in ‘Granny Smith’ and ‘Fuji’, and is negatively correlated with the acidity of the fruit [35–37]. In other studies with ‘Red Delicious’, ‘Golden Supreme’, ‘Gala’, ‘Fuji’, ‘Empire’, and ‘McIntosh’, the varieties that showed the highest patulin accumulation were ‘Golden Supreme’ (54.2 \(\mu g\ \text{kg}^{-1}\)) and ‘McIntosh’ (52.1 \(\mu g\ \text{kg}^{-1}\)) [38]. More recent indications by Snini et al. [39] suggest that patulin is not indispensable for the initiation of the disease, but it acted as a cultivar-dependent aggressiveness factor for \(P. expansum\) in 13 different cultivars tested. This conclusion was strengthened by the fact that the addition of patulin to apples infected by the
PeΔpatL mutant lacking one of the genes of the patulin production in the cluster required for patulin synthesis restored normal *P. expansum* colonization in the apples [39].

When colonization of *P. expansum* was compared in pears and apples, its growth was higher in the former; however, the latter tended to accumulate more patulin [32], suggesting that internal compounds may contribute to differential patulin accumulation. The effect of pH was also evaluated in apples: patulin production increased from pH 2.5 to 3.5 and then remained constant to pH 5.5. Other factors, such as fruit organic acid content and degree of ripeness, may play important roles in patulin accumulation [40,41].

**Storage temperature**—Optimal patulin production has been reported in the temperature range of 23–25 °C [30,32]. As the temperature decreased during storage (0–4 °C), patulin production declined but was not completely inhibited [29,30,40,42].

**Environmental factors**—Zong and co-workers [43] reported that environmental factors such as pH, and chemical factors such as carbon (C) and nitrogen (N) sources have large effects on patulin production in *P. expansum* strains. They found glutamic acid to be the best N source and sucrose, maltose, and glucose the best C sources in the promotion of patulin production, and the optimal pH was 5 in different *P. expansum* strains.

### 2.2. Ochratoxin

Ochratoxins are a group of pentaketide mycotoxins found in fruit that are produced mainly by *Aspergillus* and *Penicillium* species [44,45]. The most important and most toxic ochratoxin found naturally in foods is ochratoxin A (OTA), which shows hepatotoxicity, teratogenicity, carcinogenicity, cytotoxicity, neurotoxicity, and immunosuppressive properties [46–51]. Two other forms of ochratoxin—B and C—are less toxic and less common [52,53]. OTA is generally produced by *Aspergillus ochraceus* [54] and *Aspergillus carbonarius*, which typically infect wine grapes, and *Aspergillus alliaceus*, which contaminates nuts and figs [55,56].

**Factors Affecting OTA Production**

**Temperature and a_w**—These factors are likely to affect the rate of growth and OTA production in *Aspergillus* species in grapes. The optimal temperature for OTA production is in the reported range of 25–30 °C for *A. carbonarius*, and 30–37 °C for *A. niger* and *A. ochraceus* [57–60]. The optimal a_w for toxin production is 0.95–0.99 for *A. carbonarius* and 0.90–0.95 for *A. niger* [61]. OTA production is also strongly influenced by culture pH, with large amounts of OTA being produced at pH < 7.0 and reduced amounts at higher pH values in *A. ochraceus* [62,63].

**Fruit development conditions**—OTA accumulates to high levels during *Aspergillus* colonization of grapes with high sugar content (16–20% TSS). When grapes’ sugar content increases, they are more susceptible to infection by *A. carbonarius* and are also capable of supporting OTA production [64–66]. Delayed harvest of mature berries also increases the risk of OTA contamination [66,67]. Hot weather coupled with increased humidity and rainfall increased *Aspergillus* incidence and OTA contamination in some studies [68,69]. OTA production was correlated with severity of infection for certain varieties of grapes but not others. Nevertheless, differences in OTA contamination among varieties were often associated more strongly with seasonal variations in climate and time of ripening than with inherent characteristics of the variety [64,70].

**Storage conditions**—OTA is concentrated in berries displaying visible disease damage [65,71]. Cold storage of table grapes (0 °C) with sulfur dioxide-generating pads reduced the incidence of black *Aspergillus* species [72]. In addition, rapid drying of grapes above 30 °C to a safe a_w reduced the potential for OTA production [73].

**Nutritional factors**—The type of C source also influences OTA accumulation: glucose, sucrose, maltose, galactose, xylose and glycerol repressed OTA production, whereas the presence of lactose resulted in a ca. 7-fold increase in OTA production by *A. ochraceus* [56]. Addition of other C sources, such as arabinose, to fungal media increased OTA accumulation by *A. ochraceus, A. carbonarius*, and *A. tubingensis* [74].
N sources, specifically ammonium nitrate and acetate, also enhanced OTA production [75] compared to ammonium sulfate and chloride [56,63]. Abbas et al. [56] also indicated that other organic N sources, such as urea and glutamine, enhance OTA synthesis by A. ochraceus, and phenylalanine specifically favored OTA production in A. ochraceus, A. carbonarius, and A. tubingensis [74].

In a recent work, systematic expression analysis of OTA mycotoxin biosynthesis genes was performed to examine the relationship between growth and general expression patterns in relation to single environmental factors, such as temperature, a_{w}, and pH, and a_{w}–temperature interactions [76]. Abiotic factors such as temperature, a_{w} and pH were found to have a strong influence on the expression of mycotoxin biosynthesis genes, in agreement with the findings of several other studies [77–80]. The expression profile pattern was more pronounced in relation to changes in temperature and a_{w} than to changes in pH.

2.3. Alternaria Toxins

Alternaria species are saprophytic in nature, and widely distributed in the soil and on plant surfaces. Several species are known to grow well at low temperatures and are responsible for fruit spoilage during refrigerated transport and postharvest storage, causing severe postharvest economic losses [23,81]. Alternaria species have been reported to produce mycotoxins in fruit [82–85] such as apples, berries, oranges, tomatoes, lemons, and grapes [81,86,87]. Alternaria species are also common pathogens of tomatoes, peppers, and eggplant, in which fungal infection is generally initiated by wounds at the calyx scar [88,89]. The most important Alternaria mycotoxins can be grouped into three different structural classes: alternariol (AOH) and its monomethyl ether (AME), as well as altenuene, all dibenzopyrone derivatives; tenuazonic acid (TeA), a tetramic acid derivative, and altertoxins I, II, and III, which are perylene derivatives. The possibility that A. alternata may be a factor in several types of cancer was confirmed in several studies [90], where it was indicated that those toxins may cause cell mutagenicity and in combination with human fetal esophageal epithelium DNA, activate oncogenes [90].

Factors Affecting Alternaria Toxins

Alternaria mycotoxin production in host fruit depends upon fungal strain, host species, cultivar, and fruit maturity.

Fruit development and wounding conditions—Temperature is one of the important factors affecting the rate of colonization of Alternaria. Apples can be infected by Alternaria alternata through wounds occurring during fruit development, harvesting and storage, leading to tissue colonization and toxin accumulation. Mild temperatures (10–25 °C), relative humidity higher than 80%, and tissue susceptibility are the most strongly determinant factors for infection [6,91]. For example, Alternaria brown spot disease in citrus and persimmon fruit is more severe during conditions of rainfall and high relative humidity [92,93]. In tomatoes, host nutritional deficiencies and skin burn, together with warm and rainy weather, also enhance Alternaria infection. In naturally infected tomato fruit in Italy, the Alternaria mycotoxin TeA may reach up to 7200 µg kg^{-1} [94].

Storage conditions—Ozcelik et al. [95] reported the inhibitory effects of high-density polyethylene film packaging on Alternaria alternata colonization compared to on unpackaged tomatoes, showing a simultaneous reduction in AOH and AME and partial inhibition of fungal growth. These results indicated that differential levels of CO_{2} and O_{2}, which affect fungal development, may modulate mycotoxin production. Toxin production in synthetic media by Alternaria alternata was optimal (depending upon the type of toxin—AOH, AME, or TeA) in a temperature range of 14–28 °C [96,97]. Pose et al. [97] suggested that the most favorable temperature for AOH synthesis is 21 °C over the a_{w} range of ca. 0.922–0.982, whereas maximum AME production was at a_{w} = 0.954 and 35 °C. This could explain Hasan’s [96] finding that postharvest storage of tomato fruit at low temperature (7 °C) reduces fungal growth and toxin production.
3. Fungal and Host Regulation of Mycotoxin Synthesis in Fruit

3.1. Environmental Effects on Gene-Biosynthesis Pathways of the Fungus

As we have seen, the biosynthesis of mycotoxins is highly regulated, and substrate [98], temperature [57,99], a_w [100], and pH [101] can have profound effects. However, aside from the wide description of factors that have been reported to modulate mycotoxin production, few reports have shown the effects of environmental profiles on optimal gene activation and concurrent mycotoxin synthesis. This has been determined for OTA biosynthesis by *Penicillium verrucosum* [101–103], trichothecene biosynthesis by *Fusarium* [104–106], and aflatoxin biosynthesis by *Aspergillus* [107,108]. The recent systematic investigation of these parameters’ influence on the expression of mycotoxin biosynthesis genes [62,76,78,80,109,110] has clarified the mechanism governing external factors’ influence on the transcript expression of genes for mycotoxin biosynthesis [76]. Several other factors, such as the positive influence of oxygenic stress described on aflatoxin biosynthesis by *Aspergillus parasiticus* [111] or the application of suboptimal concentrations of some fungicides, e.g., strobilurins, can have a strong effect on mycotoxin biosynthesis [112]. Furthermore, for *P. verrucosum*, suboptimal concentrations of preservatives, such as calcium propionate and potassium sorbate, have been shown to lead to an increase in OTA biosynthesis. In parallel, it was shown that the *otapksPV* gene (encoding OTA polyketide synthase), a key gene in the OTA biosynthesis pathway, is activated under these stress conditions. This demonstrated, for the first time, that environmental factors modulate gene biosynthesis pathways [110]. Stress activation was also demonstrated for the *fum1* gene of *Fusarium verticillioides* [80].

Host Effects on the Fungus

Fruits are fresh products that undergo significant biochemical and chemical changes during postharvest storage. Fruit nutritional components that undergo changes during fruit maturation and ripening include sugars (levels may increase from 5 to 20%) and organic acids, coupled with an increase in fruit pH [79]. This raises the question of whether such changes in the host can modulate the expression of fungal mycotoxin biosynthetic gene clusters (BGCs).

With respect to host-produced metabolites, Barkai-Golan, Paster, Jackson, and Al-Taher [113,114] claimed that the high content of carbohydrates in figs, dates, citrus fruit, and raisins probably enhances aflatoxin production by *Aspergillus flavus* and *Aspergillus parasiticus*; however, no modulatory mechanism for cluster activation was indicated.

On the other hand, recent studies of patulin synthesis suggest that several host factors are involved in the inhibition of mycotoxin accumulation in apples. Patulin accumulation during colonization of apples by *Penicillium expansum* was inhibited by the addition of extracts of the mushroom *Lentinula edodes* and of the antioxidants quercetin and umbelliferone [115,116] suggesting that metabolites—produced by the host or other organisms—can alter patulin synthesis in vivo.

We envision two possible mechanisms that might modulate fungal mycotoxin cluster activation and the accumulation of mycotoxins: (1) pathogen-modified nutritional factors present in the host that become precursors for activation of the mechanism of mycotoxin production; and (2) natural chemical factors present in the host that directly modulate the transcription factors that induce mycotoxin production.

Pathogen-modified factors present in the host—Accumulation of patulin by *P. expansum* was found to be strongly dependent on the secretion of D-gluconic acid (GLA) resulting from the oxidation of host sugars by the pathogen [117]. While the acidification of host tissue contributes to the activation of pathogenicity factors and colonization, it was also found that GLA accumulation may contribute both to pH reduction of the host tissue and as a precursor for patulin [118].

Downregulation of glucose oxidase (*gox2*) in *P. expansum* was accompanied by impairment in its ability to produce GLA, patulin accumulation and apple colonization. Using *Δgox2* mutants, it was observed that the higher the impairment in GLA accumulation, the higher the inhibition of relative expression of the patulin BGC gene *idh* and patulin accumulation [118,119].
Ammonia produced by the pathogen and detected on the edge of *P. expansum*-colonized tissues under host conditions of lower sucrose level led to enhanced patulin accumulation [120]. This type of response is probably the result of reduced free sucrose levels at the leading edge of the colonized tissue inducing amino acid metabolism, in contrast to the free sugar presence at the center of the decay, where macerated tissue provides a sufficient carbon source [120]. This host effect was confirmed by direct NH₄Cl treatments to *P. expansum* colonizing fruits, where ammonia treatment induced patulin accumulation in the colonized tissue. Interestingly, ammonia induced patulin accumulation concurrently with transcript activation of *pacC* and patulin BGC genes, indicating the regulatory effect of ammonia on *pacC* transcript expression under acidic conditions [120]. These findings indicate not only that external factors affect fungal growth, but also that intrinsic metabolic changes lead to different levels of sugar availability occurring in the fruit during ripening and pathogenesis, which may affect first and secondary fungal metabolism and mycotoxin accumulation.

Another pathogen not present in fruits, for which sugar impacted toxin production, is *Stagonospora nodorum*. Deletion of the transcription factor gene *SnStuA* played a key role in regulating central C metabolism, with glycolysis, the tricarboxylic acid cycle, and amino acid synthesis in the mutants positively regulating the synthesis of the mycotoxin alternariol [121]. These data suggest that the metabolism of fermentable C sources negatively affects mycotoxin production in some cases. This study also uncovered a multitude of regulatory targets of fungal genes in plants, suggesting the possibility that other fungal–host interactions affect mycotoxin accumulation.

**Natural host factors affecting the pathogen**—Efforts to identify host factors that are potentially involved in patulin regulation have centered on the role of sucrose in impacting patulin levels. Kumar et al. [117] demonstrated that sucrose, the main sugar usually present in fruit, modulates patulin accumulation in a dose-responsive pattern by directly regulating the expression of the global regulator of secondary metabolism, *laeA*. An increase in sucrose culture amendment from 15 to 175 mM decreased both patulin accumulation and *laeA* expression by 175- and 5-fold, respectively. These results may tie into the ammonia levels, since the highest patulin accumulation was observed in the presence of low sucrose level, conditions that induce ammonia accumulation by the pathogen [122]. This suggests that limiting sugar levels, probably resulting from intact cells in unripe fruit, may be a mechanism for activating mycotoxin synthesis that differs from that in cell walls from mature ripe cells, which activates different processes of fungal metabolism. Interestingly, negative regulation of *creA* compared to *laeA* was observed at different sucrose levels, also indicating the importance of sugar regulation in these transcription factors. These data support the view that host nutritional factors, as a result of fruit maturity, may differentially contribute to regulation of the patulin BGC.

Using freshly harvested fruit sampled at increasing stages of maturity, the ability of *P. expansum* to colonize fruit and accumulate patulin was analyzed in apple fruit at increasing maturity. The wild-type strain of *P. expansum* (WT) showed increasing patulin accumulation, from 0.2 to 1.5 mg g⁻¹ fresh weight as the fruit matured, and the total soluble solids of the fruits (TSS) increased from 12.5% and 13.5% to 13.96% in the first, second, and third harvests, respectively. Thus the WT Pe-21 strain showed an increasing trend for patulin accumulation in apples in progressive harvests with progressive aggressiveness of the pathogen. The differential levels of sugar, found both in vitro and in vivo, suggest that internal metabolic factors may further contribute to the regulation of the metabolic cluster and patulin accumulation.

4. Conclusions

Changes in fruit maturity and ripening are accompanied by significant nutritional factors that determine the quality of the fruit. While many reports indicate that mycotoxin accumulation is dependent on a variety of environmental factors [5,123], there are few examples showing how changes in natural host metabolites during ripening affect fungal metabolites and the global regulation of mycotoxin synthesis. This is important given the differential levels of mycotoxin accumulation during maturation and ripening. Interestingly, until now, there have been few reports indicating the
importance of changes in the level of organic acids and other natural fruit compounds, such as phenols. In *Aspergillus*, the phenolic antioxidants gallic acid, vanillic acid, protocatechuic acid, 4-hydroxybenzoic acid, catechin, caffeic acid, and chlorogenic acid, which are present in fruit, were studied for their effects on OTA production. Whereas *Aspergillus ochraceus* was not inhibited by any of these compounds, the effects of each compound on OTA production were variable, suggesting that species-specific OTA production and response to phenolic compounds may be influenced by different and as yet unreported mechanisms affecting OTA accumulation [120].

A similar approach should be taken for organic acids, such as malic and citric acids, which are present in ripening fruit. A recent report indicates that organic acids may have antifungal activity against mycotoxigenic pathogens and an inhibitory effect on aflatoxin B1 accumulation [124]. If we take into account the dynamic changes in those natural fruit compounds and possible synergistic effects, natural host factors may regulate mycotoxin production by affecting its biosynthetic activation. Future research should focus on understanding how these natural host factors impact BGC expression and mycotoxin production during fruit colonization.

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References
1. Ueno, Y. Mode of action of trichothecenes. *Pure Appl. Chem.* 1977, 49, 1737–1745. [CrossRef]
2. Ueno, Y.; Hsieh, D.P. The toxicology of mycotoxins. *Crit. Rev. Toxicol.* 1985, 14, 99–132. [CrossRef] [PubMed]
3. European Commission. Commission regulation (EC) No. 1425/2003 of 11 August 2003 amending regulation (EC) No. 466/2001 as regards patulin. *Off. J. Eur. Union L* 2003, 203, 1–3.
4. U.S. Food and Drug Administration. Action Levels for Poisonous or Deleterious Substances in Human Food and Animal Feed. Industry Activities Staff Booklet. Food and Drug Administration August 2000. Available online: http://www.cfsan.fda.gov/_lrd/fdaact.html (accessed on 30 August 2000).
5. Drusch, S.; Ragab, W. Mycotoxins in fruits, fruit juices, and dried fruits. *J. Food Prot.* 2003, 66, 1514–1527. [CrossRef] [PubMed]
6. Sanchis, V.; Magan, N. Environmental conditions affecting mycotoxins. In *Mycotoxins in Food: Detection and Control*; Elsevier Science: Burlington, ON, Canada, 2004; Volume 103, pp. 174–189.
7. Tournas, V.; Katsoudas, E. Mould and yeast flora in fresh berries, grapes and citrus fruits. *Int. J. Food Microbiol.* 2005, 105, 11–17. [CrossRef] [PubMed]
8. Aziz, N.H.; Moussa, L.A. Influence of gamma-radiation on mycotoxin producing moulds and mycotoxins in fruits. *Food Control* 2002, 13, 281–288. [CrossRef]
9. Torres, R.; Valentines, M.; Usall, J.; Vinas, I.; Larrigaudiere, C. Possible involvement of hydrogen peroxide in the development of resistance mechanisms in ‘Golden Delicious’ apple fruit. *Postharvest Biol. Technol.* 2003, 27, 235–242. [CrossRef]
10. Monbaliu, S.; Van Pouce, C.; Van Peteghem, C.; Van Poucke, K.; Heungens, K.; De Saeger, S. Development of a multi-mycotoxin liquid chromatography/tandem mass spectrometry method for sweet pepper analysis. *Rapid Commun. Mass Spectrom.* 2009, 23, 3–11. [CrossRef] [PubMed]
11. Engelhardt, G.; Ruhland, M.; Wallnöfer, P. Metabolism of mycotoxins in plants. *Adv. Food Sci.* 1999, 21, 71–78.
12. Singh, Y.P.; Sumbali, G. Aflatoxin B1 contamination in commercial varieties of apple and pear fruits infected with *Aspergillus flavus* Link ex Fries. *Indian Phytopathol.* 2011, 64, 100–101.
13. Ciegler, A.; Vesonder, R.; Jackson, L.K. Production and biological activity of patulin and citrinin from *Penicillium expansum*. *Appl. Environ. Microbiol.* 1977, 33, 1004–1006. [PubMed]
14. Brackett, R.; Marth, E. A Research Note: Patulin in Apple Juice from Roadside Stands in Wisconsin. *J. Food Prot.* 1979, 42, 862–863. [CrossRef]

15. Hopkins, J. The toxicological hazards of patulin. *Food Chem. Toxicol. Int. J. Publ. Br. Ind. Biol. Res. Assoc.* 1993, 31, 455–456. [CrossRef] [PubMed]

16. Puel, O.; Galtier, P.; Oswald, I.P. Biosynthesis and toxicological effects of patulin. *Toxins* 2010, 2, 613–631. [CrossRef] [PubMed]

17. Scott, P.M. Collaborative study of a chromatographic method for determination of patulin in apple juice. *J. Assoc. Off. Anal. Chem.* 1974, 57, 621–625. [CrossRef]

18. Bullerman, L. Significance of mycotoxins to food safety and human health. *J. Food Prot.* 1979, 42, 65–86. [CrossRef]

19. Palmgren, M.; Ciegler, A. Toxicity and carcinogenicity of fungal lactones: Patulin and penicillic acid. *Handb. Nat. Toxins* 1983, 1, 325–341.

20. Hasan, H. Patulin and aflatoxin in brown rot lesion of apple fruits and their regulation. *World J. Microbiol. Biotechnol.* 2000, 16, 607–612. [CrossRef]

21. Snowdon, A.L. General introduction and fruits. In *A Colour Atlas of Post-Harvest Diseases and Disorders of Fruits and Vegetables*; Wolfe Scientific Ltd.: London, UK, 1990; Volume 1.

22. Larsen, T.O.; Frisvad, J.C.; Ravn, G.; Skaaning, T. Mycotoxin production by *Penicillium expansum* on blackcurrant and cherry juice. *Food Addit. Contam.* 1998, 15, 671–675. [CrossRef] [PubMed]

23. Barkai-Golan, R. *Postharvest Diseases of Fruits and Vegetables: Development and Control*; Elsevier: Amsterdam, The Netherlands, 2001.

24. Doores, S.; Splittstoesser, D.F. The microbiology of apples and apple products. *Crit. Rev. Food Sci. Nutr.* 1983, 19, 133–149. [CrossRef] [PubMed]

25. Conway, W. Effect of postharvest calcium treatment on decay of Delicious apples. *Plant Dis.* 1982, 66, 402–403. [CrossRef]

26. Nunes, C.; Usall, J.; Teixido, N.; Eribe, X.O.D.; Vinas, I. Control of post-harvest decay of apples by pre-harvest and post-harvest application of ammonium molybdate. *Pest Manag. Sci.* 2001, 57, 1093–1099. [CrossRef] [PubMed]

27. Sholberg, P.L.; Conway, W.S. Postharvest pathology. The commercial storage of fruits, vegetables, and florist and nursery stocks. In *USDA-ARS Agriculture Handbook*; United States Department of Agriculture: Washington, DC, USA, 2004.

28. Wilson, D.; Nuovo, G. Patulin production in apples decayed by *Penicillium expansum*. *Appl. Microbiol.* 1973, 26, 124–125. [PubMed]

29. Beer, S.; Amand, J. Production of the mycotoxin patulin in mature fruits of five apple cultivars infected by *Penicillium expansum*. *Proc. Am. Phytopathol. Soc.* 1974, 1, 104–110.

30. Paster, N.; Huppert, D.; Barkai-Golan, R. Production of patulin by different strains of *Penicillium expansum* in pear and apple cultivars stored at different temperatures and modified atmospheres. *Food Addit. Contam.* 1995, 12, 51–58. [CrossRef] [PubMed]

31. Martins, M.; Gimeno, A.; Martins, H.; Bernardo, F. Co-occurrence of patulin and citrinin in Portuguese apples with rotten spots. *Food Addit. Contam.* 2002, 19, 568–574. [CrossRef] [PubMed]

32. McCallum, J.; Tsao, R.; Zhou, T. Factors affecting patulin production by *Penicillium expansum*. *J. Food Prot.* 2002, 65, 1937–1942. [CrossRef] [PubMed]

33. Jackson, L.S.; Beacham-Bowden, T.; Keller, S.E.; Adhikari, C.; Taylor, K.T.; Chirtel, S.J.; Merker, R.I. Apple quality, storage, and washing treatments affect patulin levels in apple cider. *J. Food Prot.* 2003, 66, 618–624. [CrossRef] [PubMed]

34. Damoglou, A.P.; Campbell, D.S.; Button, J.E. Some factors governing the production of patulin in apples. *Food Microbiol.* 1985, 2, 3–10. [CrossRef]

35. Konstantinou, S.; Karaoglaniadis, G.; Bardas, G.; Minas, I.; Doukas, E.; Markoglou, A.N. Postharvest fruit rots of apple in Greece: Pathogen incidence and relationships between fruit quality parameters, cultivar susceptibility, and patulin production. *Plant Dis.* 2011, 95, 666–672. [CrossRef]

36. Salomão, B.C.; Aragão, G.M.; Churey, J.J.; Padilla-Zakour, O.I.; Worobo, R.W. Influence of storage temperature and apple variety on patulin production by *Penicillium expansum*. *J. Food Prot.* 2009, 72, 1030–1036. [CrossRef] [PubMed]
37. Marin, S.; Morales, H.; Hasan, H.; Ramos, A.; Sanchis, V. Patulin distribution in Fuji and Golden apples contaminated with Penicillium expansum. Food Addit. Contam. 2006, 23, 1316–1322. [CrossRef] [PubMed]
38. Pepeljnjak, S.; Šegvic, M.; Ozegovic, L. Citrininoxigenicity of Penicillium spp. isolated from decaying apples. Braz. J. Microbiol. 2002, 33, 134–137. [CrossRef]
39. Snini, S.P.; Tannous, J.; Heuillard, P.; Bailly, S.; Lippi, Y.; Zehraoui, E.; Barreau, C.; Oswald, I.P.; Puel, O. The patulin is a cultivar dependent aggressiveness factor favoring the colonization of apples by Penicillium expansum. Mol. Plant Pathol. 2016, 17, 920–930. [CrossRef] [PubMed]
40. Morales, H.; Marin, S.; Rovira, A.; Ramos, A.; Sanchis, V. Patulin accumulation in apples by Penicillium expansum during postharvest stages. Lett. Appl. Microbiol. 2007, 44, 30–35. [CrossRef] [PubMed]
41. Morales, H.; Barros, G.; Marin, S.; Chulze, S.; Ramos, A.J.; Sanchis, V. Effects of apple and pear varieties and pH on patulin accumulation by Penicillium expansum. J. Sci. Food Agric. 2008, 88, 2738–2743. [CrossRef]
42. Barkai-Golan, R. Postharvest disease suppression by atmospheric modifications. In Food Preservation by Modified Atmospheres; Calderon, M., Barkai-Golan, R., Eds.; CRC Press: Boca Raton, FL, USA, 1990; pp. 237–264.
43. Zong, Y.; Li, B.; Tian, S. Effects of carbon, nitrogen and ambient pH on patulin production and related gene expression in Penicillium expansum. Int. J. Food Microbiol. 2015, 206, 102–108. [CrossRef] [PubMed]
44. Bayman, P.; Baker, J.L. Ochratoxins: A global perspective. Mycopathologia 2006, 162, 215–223. [CrossRef] [PubMed]
45. Cabañes, F.J.; Bragulat, M.R.; Castellá, G. Ochratoxin A producing species in the genus Penicillium. Toxins 2010, 2, 1111–1120.
46. Woo, C.S.J.; Partanen, H.; Myllynen, P.; Vähäkangas, K.; El-Nezami, H. Fate of the teratogenic and carcinogenic ochratoxin A in human perfused placenta. Toxicol. Lett. 2012, 208, 92–99. [CrossRef] [PubMed]
47. Stachurska, A.; Ciesla, M.; Kozakowska, M.; Wolffram, S.; Boesch-Saadatmandi, C.; Rimbach, G.; Jozkowicz, A.; Dulak, J.; Loboda, A. Cross-talk between microRNAs, nuclear factor E2-related factor 2, and heme oxygenase-1 in ochratoxin A-induced toxic effects in renal proximal tubular epithelial cells. Mol. Nutr. Food Res. 2013, 57, 504–515. [CrossRef] [PubMed]
48. Solcan, C.; Floristean, V.; Pavel, G.; Solcan, G. Induced malabsorbtion in chickens by experimental administration of ochratoxin A. Curr. Opin. Biotechnol. 2013, 24, S105. [CrossRef]
49. Von Tobel, J.S.; Antinori, P.; Zurich, M-G.; Rosset, R.; Aschner, M.; Glück, F.; Scherl, A.; Monnet-Tschudi, F. Repeated exposure to Ochratoxin A generates a neuroinflammatory response, characterized by neurodegenerative M1 microglial phenotype. Neurotoxicology 2014, 44, 61–70. [CrossRef] [PubMed]
50. Gayathri, L.; Dhiya, R.; Dhanasekaran, D.; Periasamy, V.S.; Alshatwi, A.A.; Akbarsha, M.A. Hepatotoxic effect of ochratoxin A and citrinin, alone and in combination, and protective effect of vitamin E: In vitro study in HepG2 cell. Food Chem. Toxicol. 2015, 83, 151–163. [CrossRef] [PubMed]
51. Calado, T.; Verde, S.C.; Abrunhosa, L.; Fernández-Cruz, M.; Venâncio, A. Cytotoxicity of mycotoxins after gamma irradiation. In Proceedings of the International Conference on Food Contaminants: Challenges in Chemical Mixtures, Lisbon, Portugal, 13–14 April 2015; pp. 149–150.
52. Stormer, F. Ochratoxin A: A mycotoxin of concern. In Handbook of Applied Mycology; Elsevier Ireland limited: Dublin, Ireland, 1992; Volume 5, pp. 403–432.
53. Heussner, A.H.; Bingle, L.E. Comparative ochratoxin toxicity: A review of the available data. Toxins 2015, 7, 4253–4282. [CrossRef] [PubMed]
54. Sweeney, M.J.; Dobson, A.D. Mycotoxin production by Aspergillus, Fusarium and Penicillium species. Int. J. Food Microbiol. 1998, 43, 141–158. [CrossRef]
55. Bayman, P.; Baker, J.L.; Doster, M.A.; Michailides, T.J.; Mahoney, N.E. Ochratoxin production by the Aspergillus ochraceus group and Aspergillus alliaceus. Appl. Environ. Microbiol. 2002, 68, 2326–2329. [CrossRef] [PubMed]
56. Abbas, A.; Valez, H.; Dobson, A.D. Analysis of the effect of nutritional factors on OTA and OTB biosynthesis and polyketide synthase gene expression in Aspergillus ochraceus. Int. J. Food Microbiol. 2009, 135, 22–27. [CrossRef] [PubMed]
57. Belli, N.; Marn, S.; Sanchis, V.; Ramos, A. Influence of water activity and temperature on growth of isolates of Aspergillus section Nigri obtained from grapes. Int. J. Food Microbiol. 2004, 96, 19–27. [CrossRef] [PubMed]
58. Ayerst, G. The effects of moisture and temperature on growth and spore germination in some fungi. J. Stored Prod. Res. 1969, 5, 127–141. [CrossRef]
59. Marín, S.; Sanchis, V.; Ramos, A.; Magan, N. Environmental factors, interspecific interactions, and niche overlap between Fusarium moniliforme and F. proliferatum and Fusarium graminearum, Aspergillus and Penicillium spp. isolated from maize. Mycol. Res. 1998, 102, 831–837.

60. Su-lin, L.L.; Hocking, A.D.; Scott, E.S. Effect of temperature and water activity on growth and ochratoxin A production by Australian Aspergillus carbonarius and A. niger isolates on a simulated grape juice medium. Int. J. Food Microbiol. 2006, 110, 209–216.

61. Selouane, A.; Bouya, D.; Lebrithi, A.; Decock, C.; Bouseta, A. Impact of some environmental factors on growth and production of ochratoxin A of/by Aspergillus tubingensis, A. niger, and A. carbonarius isolated from Moroccan grapes. J. Microbiol. 2009, 47, 411–419. [CrossRef][PubMed]

62. O’Callaghan, J.; Stapleton, P.C.; Dobson, A.D. Ochratoxin A biosynthetic genes in Aspergillus ochraceus are differentially regulated by pH and nutritional stimuli. Fungal Genet. Biol. 2006, 43, 213–221.

63. Mühlencoert, E.; Mayer, I.; Zapf, M.W.; Vogel, R.F.; Niessen, L. Production of ochratoxin A by Aspergillus ochraceus. In Molecular Diversity and PCR-Detection of Toxigenic Fusarium Species and Ochratoxigenic Fungi; Springer: Dordrecht, The Netherlands, 2004; pp. 651–659.

64. Battilani, P.; Barbano, C.; Marin, S.; Sanchis, V.; Kozakiewicz, Z.; Magan, N. Mapping of Aspergillus section Nigri in Southern Europe and Israel based on geostatistical analysis. Int. J. Food Microbiol. 2006, 111, S72–S82. [CrossRef][PubMed]

65. Leong, S.L.; Hien, L.T.; An, T.V.; Trang, N.T.; Hocking, A.D.; Scott, E.S. Ochratoxin A-producing Aspergillus in Vietnamese green coffee beans. Lett. Appl. Microbiol. 2007, 45, 301–306. [CrossRef][PubMed]

66. Roset, M. Quality control survey. On ochratoxin a in grape juice. Fruit processing. J. Fruit Process. Juice Prod. Eur. Overseas Ind. 2003, 13, 167–172.

67. Gambuti, A.; Strollo, D.; Genovese, A.; Ugliano, M.; Ritienni, A.; Moio, L.; Vitic, A.J.E. Influence of enological practices on ochratoxin A concentration in wine. Am. J. Enol. Vitic. 2005, 56, 155–162.

68. Battilani, P.; Giorni, P.; Pietri, A. Epidemiology of toxin-producing fungi and ochratoxin A occurrence in grapes. J. Plant Pathol. 2003, 109, 715–722. [CrossRef]

69. Bell, N.; Mitchell, D.; Marín, S.; Alegre, I.; Ramos, A.J.; Magan, N.; Sanchis, V. Ochratoxin A-producing fungi in Spanish wine grapes and their relationship with meteorological conditions. Eur. J. Plant Pathol. 2005, 113, 233–239. [CrossRef]

70. Leong, S.L.; Hocking, A.D.; Pitt, J.I.; Kazi, B.A.; Emmett, R.W.; Scott, E.S. Australian research on ochratoxigenic fungi and ochratoxin A. Int. J. Food Microbiol. 2006, 111, S10–S17. [CrossRef][PubMed]

71. Guzev, L.; Danshin, A.; Ziv, S.; Lichter, A. Occurrence of ochratoxin A producing fungi in wine and table grapes in Israel. Int. J. Food Microbiol. 2006, 111, S67–S71. [CrossRef][PubMed]

72. Lichter, A.; Danshin, A.; Zahavi, T.; Ovadia, A.; Guzev, L. Survival of OTA producing fungi during storage of table grapes. Abstracts, Ochratoxin A in Grapes and Wine: Prevention and Control 2005.

73. Hocking, A.D.; Su-lin, L.L.; Kazi, B.A.; Emmett, R.W.; Scott, E.S. Fungi and mycotoxins in vineyards and grape products. Int. J. Food Microbiol. 2007, 119, 84–88. [CrossRef][PubMed]

74. Medina, A.; Mateo, E.M.; Valle-Algarra, F.M.; Mateo, F.; Mateo, R.; Jiménez, M. Influence of nitrogen and carbon sources on the production of Ochratoxin A by ochratoxigenic strains of Aspergillus spp. isolated from grapes. Int. J. Food Microbiol. 2008, 122, 93–99. [CrossRef][PubMed]

75. Ferreira, N.; Pitout, M. Biogenesis of ochratoxin. J. S. Afr. Chem. Inst. 1969, 22, S1.

76. Schmidt-Heydt, M.; Magan, N.; Geisen, R. Stress induction of mycotoxin biosynthesis genes by abiotic factors. FEMS Microbiol. Lett. 2008, 284, 142–149. [CrossRef][PubMed]

77. Keller, N.P.; Hohn, T.M. Metabolic pathway gene clusters in filamentous fungi. Fungal Genet. Biol. 1997, 21, 17–29. [CrossRef][PubMed]

78. Geisen, R.; Mayer, Z.; Karolewicz, A.; Färber, P. Development of a real time PCR system for detection of Penicillium nordicum and for monitoring ochratoxin A production in foods by targeting the ochratoxin polyketide synthase gene. Syst. Appl. Microbiol. 2004, 27, 501–507. [CrossRef][PubMed]

79. Price, M.S.; Connors, S.B.; Tachdjian, S.; Kelly, R.M.; Payne, G.A. Aflatoxin conducive and non-conducive growth conditions reveal new gene associations with aflatoxin production. Fungal Genet. Biol. 2005, 42, 506–518. [CrossRef][PubMed]

80. Jurado, M.; Marín, P.; Magan, N.; González-Jaén, M.T. Relationship between oolute and matric potential stress, temperature, growth, and FUM1 gene expression in two Fusarium verticillioides Strains from Spain. Appl. Environ. Microbiol. 2008, 74, 2032–2036. [CrossRef][PubMed]
81. Stinson, E.E.; Osman, S.F.; Heisler, E.G. Mycotoxin production by Alternaria species grown on apples, tomatoes, and blueberries. J. Agric. Food Chem. 1980, 28, 960–963. [CrossRef] [PubMed]
82. Scott, P.; Kanhere, S. Liquid chromatographic determination of tenuazonic acids in tomato paste. J. Assoc. Off. Anal. Chem. 1980, 63, 612–621. [PubMed]
83. Stack, M.E.; Mislivec, P.B.; Roach, J.; Pohland, A.E. Liquid chromatographic determination of tenuazonic acid and alternariol methyl ether in tomatoes and tomato products. J. Assoc. Off. Anal. Chem. 1984, 68, 640–642.
84. Fente, C.; Jaimez, J.; Vázquez, B.; Franco, C.; Cepeda, A. Determination of alternariol in tomato paste using solid phase extraction and high-performance liquid chromatography with fluorescence detection. Analyst 1998, 123, 2277–2280. [CrossRef] [PubMed]
85. Da Motta, S.; Valente Soares, L.M. Survey of Brazilian tomato products for alternariol, alternariol monomethyl ether, tenuazonic acid and cyclopiazonic acid. Food Addit. Contam. 2001, 18, 630–634. [CrossRef] [PubMed]
86. Stinson, E.E.; Osman, S.F.; Heisler, E.G.; Siciliano, J.; Bills, D.D. Mycotoxin production in whole tomatoes, apples, oranges, and lemons. J. Agric. Food Chem. 1981, 29, 790–792. [CrossRef] [PubMed]
87. Tournas, V.; Stack, M.E. Production of alternariol and alternariol methyl ether by Alternaria alternata grown on fruits at various temperatures. J. Food Prot. 2001, 64, 528–532. [CrossRef] [PubMed]
88. Dennis, C. Post-Harvest Pathology of Fruits and Vegetables: JSTOR; Academic Press: New York, NY, USA, 1983.
89. Fallik, E.; Aharoni, Y.; Grinberg, S.; Copel, A.; Klein, J. Postharvest hydrogen peroxide treatment inhibits decay in eggplant and sweet red pepper. Crop Prot. 1994, 13, 451–454. [CrossRef]
90. Ostry, V. Alternaria mycotoxins: An overview of chemical characterization, producers, toxicity, analysis and occurrence in foodstuffs. World Mycotoxin J. 2008, 1, 175–188. [CrossRef]
91. Reuveni, M.; Sheglov, D.; Sheglov, N.; Ben-Arie, R.; Prusky, D. Sensitivity of Red Delicious apple fruit at various phenologic stages to infection by Alternaria alternata and moldy-core control. Eur. J. Plant Pathol. 2002, 108, 421–427. [CrossRef]
92. Timmer, L.; Solé, Z.; Gottwald, T.; Ibanez, A.; Zitko, S. Environmental factors affecting production, release, and field populations of conidia of Alternaria alternata, the cause of brown spot of citrus. Phytopathology 1998, 88, 1218–1223. [CrossRef] [PubMed]
93. Prusky, D.; Fuchs, Y.; Yanko, U. Assessment of latent infections as a basis for control of postharvest disease of mango. Plant Dis. 1983, 67, 816–818. [CrossRef]
94. Panigrahi, S. Alternaria Toxins. Handbook of Plant and Fungal Toxicants, 1st ed.; CRC Press: Boca Raton, FL, USA, 1997; pp. 319–337.
95. Ozcelik, S.; Ozcelik, N.; Beuchat, L.R. Toxin production by Alternaria alternata in tomatoes and apples stored under various conditions and quantitation of the toxins by high-performance liquid chromatography. Int. J. Food Microbiol. 1990, 11, 187–194. [CrossRef]
96. Hassan, H. Alternaria mycotoxins in Black rot lesion of tomato fruits: Conditions and regulations of their productions. Acta Immunol. Hung. 1996, 43, 125–133. [CrossRef]
97. Pose, G.; Patriarca, A.; Kyanko, V.; Pardo, A.; Pinto, V.F. Water activity and temperature effects on mycotoxin production by Alternaria alternata in a synthetic tomato medium. Int. J. Food Microbiol. 2010, 142, 348–353. [CrossRef] [PubMed]
98. Skrinjar, M.; Dimic, G. Ochratoxigenicity of Aspergillus ochraceus group and Penicillium verrucosum var. cyclopium strains on various media. Acta Microbiol. Hung. 1992, 39, 257–261. [PubMed]
99. Mitchell, D.; Parra, R.; Aldred, D.; Magan, N. Water and temperature relations of growth and ochratoxin A production by Aspergillus carbonarius strains from grapes in Europe and Israel. J. Appl. Microbiol. 2004, 97, 439–445. [CrossRef] [PubMed]
100. Valero, A.; Farré, J.R.; Sanchis, V.; Ramos, A.J.; Marin, S. Effects of fungal interaction on ochratoxin A production by A. carbonarius at different temperatures and aw. Int. J. Food Microbiol. 2006, 110, 160–164. [CrossRef] [PubMed]
101. Arroyo, M.; Aldred, D.; Magan, N. Environmental factors and weak organic acid interactions have differential effects on control of growth and ochratoxin A production by Penicillium verrucosum isolates in bread. Int. J. Food Microbiol. 2005, 98, 223–231. [CrossRef] [PubMed]
102. Cairns-Fuller, V.; Aldred, D.; Magan, N. Water, temperature and gas composition interactions affect growth and ochratoxin A production by isolates of Penicillium verrucosum on wheat grain. J. Appl. Microbiol. 2005, 99, 1215–1221. [CrossRef] [PubMed]
103. Pardo, E.; Malet, M.; Marin, S.; Sanchis, V.; Ramos, A. Effects of water activity and temperature on germination and growth profiles of ochratoxigenic Penicillium verrucosum isolates on barley meal extract agar. Int. J. Food Microbiol. 2006, 106, 25–31. [CrossRef] [PubMed]

104. Llorens, A.; Mateo, R.; Hinojo, M.; Valle-Algarra, F.; Jimenez, M. Influence of environmental factors on the biosynthesis of type B trichothecenes by isolates of Fusarium spp. from Spanish crops. Int. J. Food Microbiol. 2004, 94, 43–54. [CrossRef] [PubMed]

105. Hope, R.; Aldred, D.; Magan, N. Comparison of environmental profiles for growth and deoxynivalenol production by Fusarium culmorum and F. graminearum on wheat grain. Lett. Appl. Microbiol. 2005, 40, 295–300. [CrossRef] [PubMed]

106. Ramirez, M.L.; Chulze, S.; Magan, N. Temperature and water activity effects on growth and temporal deoxynivalenol production by two Argentinean strains of Fusarium graminearum on irradiated wheat grain. Int. J. Food Microbiol. 2006, 106, 291–296. [CrossRef] [PubMed]

107. Nesci, A.; Rodriguez, M.; Etcheverry, M. Control of Aspergillus growth and aflatoxin production using antioxidants at different conditions of water activity and pH. J. Appl. Microbiol. 2003, 95, 279–287. [CrossRef] [PubMed]

108. Ribeiro, J.M.M.; Cavaglieri, L.R.; Fraga, M.E.; Direito, G.M.; Dalcero, A.M.; Rosa, C.A.R. Influence of water activity, temperature and time on mycotoxins production on barley rootlets. Lett. Appl. Microbiol. 2006, 42, 179–184. [CrossRef] [PubMed]

109. Feng, G.H.; Leonard, T.J. Culture conditions control expression of the genes for aflatoxin and sterigmatocystin production by Aspergillus parasiticus and A. nidulans. Appl. Environ. Microbiol. 1998, 64, 2275–2277. [PubMed]

110. Schmidt-Heydt, M.; Geisen, R. A microarray for monitoring the production of mycotoxins in food. J. Appl. Microbiol. 2007, 102, 225–237. [CrossRef] [PubMed]

111. Jayashree, T.; Subramanyam, C. Oxidative stress as a prerequisite for aflatoxin production by Aspergillus parasiticus. Free Radic. Biol. Med. 2000, 29, 981–985. [CrossRef] [PubMed]

112. Ellner, F. Results of long-term field studies into the effect of strobilurin containing fungicides on the production of mycotoxins in several winter wheat varieties. Mycotoxin Res. 2005, 21, 112–115. [CrossRef] [PubMed]

113. Barkai-Golan, R.; Paster, N. Mouldy fruits and vegetables as a source of mycotoxins: Part 1. World Mycotoxin J. 2008, 1, 147–159. [CrossRef]

114. Jackson, L.S.; Al-Taher, F. Factors Affecting Mycotoxin Production in Fruits; Elsevier: San Diego, CA, USA, 2008.

115. Sanzani, S.M.; Schena, L.; Nigro, F.; De Girolamo, A.; Ippolito, A. Effect of quercetin and umbelliferone on the transcript level of Penicillium expansum genes involved in patulin biosynthesis. Eur. J. Plant Pathol. 2009, 125, 223–233. [CrossRef] [PubMed]

116. Tolaini, V.; Zajlic, S.; Reverberi, M.; Fanelli, C.; Fabbri, A.A.; Fiore, A.D.; Rossi, P.D.; Ricelli, A. Lentinula edodes enhances the biocontrol activity of Cryptococcus laurentii against Penicillium expansum contamination and patulin production in apple fruits. Int. J. Food Microbiol. 2010, 138, 243–249. [CrossRef] [PubMed]

117. Kumar, D.; Barad, S.; Chen, Y.; Luo, X.; Tannous, J.; Dubey, A.; Matana, N.G.; Tian, S.; Li, B.; Keller, N.; et al. LaeA regulation of secondary metabolism modulates virulence in Penicillium expansum and is mediated by sucrase. Mol. Plant Pathol. 2016. [CrossRef] [PubMed]

118. Barad, S.; Horowitz, S.B.; Kobiler, I.; Sherman, A.; Prusky, D. Accumulation of the mycotoxin patulin in the presence of gluconic acid contributes to pathogenicity of Penicillium expansum. Mol. Plant-Microbe Interact. 2014, 27, 66–77. [CrossRef] [PubMed]

119. Barad, S.; Horowitz, S.B.; Moscovitz, O.; Lichter, A.; Sherman, A.; Prusky, D. A Penicillium expansum glucose oxidase–encoding gene, GOX2, is essential for gluconic acid production and acidification during colonization of deciduous fruit. Mol. Plant-Microbe Interact. 2012, 25, 779–788. [CrossRef] [PubMed]

120. Palumbo, J.D.; O’Keeffe, T.L.; Mahoney, N.E. Inhibition of ochratoxin A production and growth of Aspergillus species by phenolic antioxidant compounds. Mycopathologia 2007, 164, 241–248. [CrossRef] [PubMed]

121. Chooi, Y.H.; Muria-Gonzalez, M.J.; Mead, O.L.; Solomon, P.S. SnPKS19 encodes the polyketide synthase for alternariol mycotoxin biosynthesis in the wheat pathogen Parastagonospora nodorum. Appl. Environ. Microbiol. 2015, 81, 5309–5317. [CrossRef] [PubMed]
122. Barad, S.; Espeso, E.A.; Sherman, A.; Prusky, D. Ammonia activates pacC and patulin accumulation in an acidic environment during apple colonization by *Penicillium expansum*. *Mol. Plant Pathol.* 2016, 17, 727–740. [CrossRef] [PubMed]

123. Magan, N.; Cayley, G.R.; Lacey, J. Effect of water activity and temperature on mycotoxin production by *Alternaria alternata* in culture and on wheat grain. *Appl. Environ. Microbiol.* 1984, 47, 1113–1117. [PubMed]

124. Hassan, A.M.; Abdel-Aziem, S.H.; El-Nekeety, A.A.; Abdel-Wahhab, M.A. Panax ginseng extract modulates oxidative stress, DNA fragmentation and up-regulate gene expression in rats sub chronically treated with aflatoxin B1 and fumonisin B1. *Cytotechnology* 2015, 67, 861–871. [CrossRef] [PubMed]

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