## Energy and Nutrients' Recovery from Contaminated Food Products

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Mycotoxins' contamination of food products is a well-known issue that is gaining interest nowadays due to increasing contaminations that are also related to climate change. Considering the principles of Circular Economy, finding robust and reliable strategies for the decontamination and valorisation of mycotoxin-contaminated products becomes mandatory. Anaerobic digestion (AD) and composting appear as promising biological treatments to degrade mycotoxins and allow for recovering energy (i.e., biogas production) and materials (i.e., nutrients from digestate and/or compost).



# **1.** Biological Treatments for Energy and Nutrients' Recovery from Organic Wastes

Anaerobic digestion (AD) and composting are the most widespread biological systems used for organic waste treatments. These technologies allow for recovering energy and nutrients from livestock residues, sludge, the agroindustrial, municipal and food wastes, making their disposal more sustainable in both economic and environmental perspectives.

AD and composting are two distinct biological processes, where organic matter is biodegraded under anaerobic and aerobic conditions, respectively. AD can degrade organic wastes to biogas and digestate through four subsequent phases, (1) hydrolysis, (2) acidogenesis, (3) acetogenesis, and (4) methanogenesis <sup>[1]</sup>. Biogas is a gas mixture mainly composed by methane and carbon dioxide (55–70% and 30–45%), as well as small amounts of other gases (oxygen, sulphuric acid, and hydrogen) <sup>[2]</sup>. Biogas can be used as an alternative energy source through its combustion in boilers or combined heat and power units; however, the interest in biogas conversion to high-value products is increasing recently <sup>[2][3]</sup>. Digestate is widely considered as a potential organic fertilizer, being rich in plant macronutrients (N, P and K) and organic matter <sup>[4][5][6]</sup>. Ammonium-N is the most abundant form of N in digestate, making digestate a readily available N fertilizer for plants <sup>[4][6]</sup>. Concerns emerged regarding organic matter stabilization in digestate <sup>[7][8][9]</sup>, but Tambone et al. <sup>[10]</sup> stated that the well-performed AD can produce stabilized digestate due to the mineralization of labile organic compounds and persistence of recalcitrant organic molecules. Nevertheless, digestate often requires post-treatments to improve its agricultural reuse (i.e., to reduce water content). AD can be operated at psychrophilic (18–20 °C), mesophilic (35–40 °C) and thermophilic (50–60

°C) temperature regimes <sup>[11]</sup>, the last two conditions being more effective for organic matter degradation and biogas production. Besides some disadvantages (e.g., slow degradation, need for post-treatment of digestate, process instability), AD is characterized by several important advantages such as net energy production, reduced odor, and small area requirement <sup>[1][12]</sup>.

Composting degrades aerobically organic wastes to compost and two main by-products (heat and carbon dioxide) <sup>[13]</sup>. Composting is a self-heating process that proceeds through three main phases, (1) mesophilic (25–40 °C), (2) thermophilic (55–65 °C) and (3) cooling and maturation. Compost is defined as a nutrient-rich organic amendment able to provide N, P, K, and organic matter to soil <sup>[14]</sup>. Differently from digestate, organic-N is predominant in compost, making it a long-term N source for plants <sup>[15]</sup>. During composting, labile organic matter is mineralized, and complex recalcitrant materials tend to concentrate, conferring stability to the compost <sup>[15]</sup>. Compost also positively affects physical properties of soil (e.g., porosity and water-holding capacity) mainly due to its reduced bulk density <sup>[17]</sup>. Lin et al. <sup>[1]</sup> reviewed the composting of organic wastes and indicated the fast degradation, small investments, and compost reuse as the main advantages of this process, the gas emissions, large area requirements and the net energy consumption being its main disadvantages.

Although AD and composting can effectively reduce environmental and financial costs of organic waste disposal, the possible contamination of soil, water and air related to the agricultural reuse of digestate, and compost emerged recently as a critical issue. In fact, the environment contamination derived by soil fertilization with waste-derived fertilizers can rise risks to human, animal, and plant health <sup>[18]</sup>. A large set of emerging contaminants can occur in organic wastes (e.g., antimicrobials, antimicrobial resistance genes, pesticides, heavy metals) and their fate during AD, and composting was recently reviewed by Congilosi and Aga <sup>[19]</sup>. Sertillanges et al. <sup>[20]</sup> evaluated the fate of organic micro-pollutants during the industrial scale treatment of organic wastes and observed that process type and compound characteristics mainly influence the pollutants' fate, the waste origin not being significant. For instance, the antimicrobials demonstrated a variable fate during AD and composting. Some studies stated that various antimicrobials degraded completely, whereas others reported only a partial degradation <sup>[19][21]</sup> [<sup>22][23]</sup>. Similar results have been reported for other emerging pollutants such as antimicrobial resistance genes, hormones, and pesticides <sup>[24][25][26]</sup>. It seems that the synergic effects of microbial activity, temperature, pH, binding to organic matrix, and mineralization during AD and composting contribute to organic pollutants' degradation <sup>[19]</sup>. Concerning the threat of heavy metals, AD and composting cannot degrade them but biological treatments are known to reduce their bioavailability <sup>[27]</sup>.

### 2. AD of Contaminated Food Products

AD of AF-contaminated matrices for energy and nutrients' recovery has been investigated only in the last decade. To have a complete view of the topic of mycotoxins' effects on AD, the present review includes also papers dealing with mycotoxins different from AFs.

Recent papers investigated the effect of mycotoxins on biogas production and digestate quality using both batch and CSTR (continuous-stirred tank reactor) trials, as well as mycotoxins' fate during the anaerobic process (**Table**  1 and Table 2).

Mycotoxin	Anaerobic Digestion	Organic Substrate	Biogas Production (NL/kg TS)	Methane (% v/v)	Process Stability I	References
	Batch mesophilic	Corn grain	579–617	57–60	n.a.	
AFB1	CSTR mesophilic	Corn grain	580	58	VFA, VFA/alkalinity, ammonium-N in optimal range	[ <u>28]</u>
AFB1	CSTR mesophilic	Corn flour	600–625	50–55	VFA, VFA/alkalinity, pH in optimal range	[ <u>29]</u>
FB1 + FB2 + FB3	Batch mesophilic	Corn silage	170–180	55	pH in optimal range	[ <u>30]</u>
	Batch mesophilic	Wholewheat flour	340	55	n.a.	
	Batch mesophilic	Wheat bran	330	55	n.a.	
DON + T-2 + HT-2	Batch mesophilic	Wheat fine bran	350	55	n.a.	[ <u>31</u> ]
	Batch mesophilic	Wheat semolina	350	50	n.a.	
	Batch mesophilic	Wheat fine middlings	300	50	n.a.	
AFB1 + DON + ZEN + OTA +	Batch mesophilic	Corn grain	500-550	55–60	n.a.	
ergot alkaloid mix	Batch thermophilic	Corn grain	580–620	55–60	n.a.	
DON + 3- ADON + 15- ADON + AOH + T-2 + ZEN + FB1 + FB2 + ENNB	CSTR mesophilic	Corn grain	680	60–65	VFA, VFA/alkalinity, pH in optimal range	[ <u>32</u> ]

#### Table 1. Biogas production and process stability.

Mycotoxin	Anaerobic Digestion	Organic Substrate	Biogas Production (NL/kg TS)	Methane (% v/v)	Process Stability	References
DON	Batch mesophilic	Wheat flour	667.2–742.8	50–55	n.a.	[ <u>33]</u>
	CSTR thermophilic	Corn grain	690	60–65	VFA, VFA/alkalinity, pH in optimal range	
AFB1	CSTR mesophilic	Corn grain	700–800 (25 μg kg <sup>-1</sup> AFB1)	60–65	VFA, VFA/alkalinity, ammonium-N, and pH in optimal range	
	CSTR mesophilic	Corn grain	0 (100 μg kg <sup>-1</sup> AFB1)	0	VFA accumulation and pH decrease to inhibiting values	[34]
AFB1 + DON + ZEN + OTA +	Batch mesophilic	Corn grain	500–550	55–60	n.a.	
ergot alkaloid mix	Batch thermophilic	Corn grain	580–620	55–60	n.a.	
DON + 3- ADON + 15-	CSTR mesophilic	Corn grain	680	60–65	VFA, VFA/alkalinity, pH in optimal range	
Mycotoxin	Initial Contan kg⁻	nination (µg <sup>·1</sup> )	Anaerobic Digestion	Organi Substra	ic Average Ate Mycotoxin Removal	References
AER1	0.54–1	.10.0	Batch mesophilic	Corn gra	ain 69–87%	[ <u>28]</u>
	7.2	2	CSTR mesophilic	Corn gra	ain 61%	
AFB1	2–4	70	CSTR mesophilic	Corn flo	ur 12–95%	[29]
FB1 + FB2 + FB3 + AFB1	241.5–13874 (FB1) + 866.5–3877 (FB2) + 42.5– 3591 (FB3) + 251 (AFB1)		Batch mesophilic	Corn sila	20–60% (FB1, FB2, FB3) 55% (AFB1)	[ <u>30]</u>
DON + T-2 + HT- 2	368–12,916 (E (T-2+H	DON) + 5–65 1T-2)	Batch mesophilic	Wholewh flour	89.9% eat (DON) 100% (T-2, HT-2)	[ <u>31</u> ]
	368–12,916 (C (T-2 + )	DON) + 5–65 HT-2)	Batch mesophilic	Wheat br	an 88.5% (DON)	

Mycotoxin	Initial Contamination (µg kg <sup>-1</sup> )	Anaerobic Digestion	Organic Substrate	Average Mycotoxin Removal	References
				100% (T-2, HT-2)	
	368–12,916 (DON) + 5–65 (T-2 + HT-2)	Batch mesophilic	Wheat fine bran	83.9% (DON) 100% (T-2, HT-2)	
	368–12,916 (DON) + 5–65 (T-2 + HT-2)	Batch mesophilic	Wheat semolina	82.1% (DON) 100% (T-2, HT-2)	
	368–12,916 (DON) + 5–65 (T-2 + HT-2)	Batch mesophilic	Wheat fine middlings	98.7% (DON) 100% (T-2, HT-2)	
AFB1 + DON + ZEN + OTA + FB1 + T-2 + ergot alkaloid mix	40 (AFB1) + 300 (DON) + 100 (ZEN) + 50 (OTA) + 100 (FB1) + 100 (T-2) + 40 (ergot alkaloid mix)	Batch mesophilic	Corn grain	>90% (AFB1, DON, ZEN, OTA, T-2) 70% (FB1) 64% (ergot alkaloid mix)	
		Batch thermophilic	Corn grain	>90% (AFB1, DON, ZEN, OTA, T-2) 85% (FB1) 98% (ergot alkaloid mix)	[ <u>32</u> ]
DON + 3-ADON + 15-ADON +	4413 (DON) + 729 (3- ADON + 15-ADON) + 14	CSTR mesophilic	Corn grain	>99%	
ZEN + FB1 + FB2 + ENNB	(AOH) + 28 (1-2) + 1052 (ZEN) + >80 (FB1 + FB2) + >80 (ENNB)	CSTR thermophilic	Corn grain	>99%	
DON	1976-80,000	Batch mesophilic	Wheat flour	100%	[ <u>33]</u>
	25	CSTR mesophilic	Corn grain	18.8% *	[ <u>34]</u>
AFB1	100	CSTR mesophilic	Corn grain	37.2% * [ <u>28</u> ]	

out both batch and continuous-stirred tank reactor (CSTR) experiments fed with pig slurry and AFB1-contaminated corn. In batch tests, a stable methane production (57–60% v/v) was achieved in all the trials, even when the highest concentration of AFB1 was tested (110 µg kg corn<sup>-1</sup> wet weight). The cumulative biogas production was in accordance with values reported in literature for corn grain (350–375 NL kgTS<sup>-1</sup>). CSTR experiments were

operated with 40 days of HRT through the dail**g**cadditidation to AFBS h contaminated feedstock to reproduce a fullscale digestion process. Biogas production and chemical parameters (e.g., volatile fatty acids, ammonia content) of the CSTR experiments did not demonstrate differences between non-contaminated and contaminated tests. Salati et al. <sup>[27]</sup> concluded that AFB1 did not affect the AD of pig slurry and corn grain. These results were in accordance with Giorni et al. <sup>[29]</sup>, who demonstrated that mycotoxins do not affect biogas production from the AD of cattle manure, corn silage and corn flour. They studied two different levels of AFB1 and fumosin contamination (70 and 470 μg kg flour<sup>-1</sup> AFB1 and 1200 and 3700 μg kg flour<sup>-1</sup> fumosins) using CSTR reactors operating with an HRT of 50 days. Biogas quantity and quality were not affected by mycotoxins, as well as process stability (FOS/TAC ratio was in the optimal range throughout the experiments).

Other studies support the conclusion that biogas can be effectively recovered from mycotoxin-contaminated matrices <sup>[30][31][32][33]</sup>. When batch AD tests carried out using corn as feedstock were spiked with 40  $\mu$ g kg<sup>-1</sup> AFB1, 300  $\mu$ g kg<sup>-1</sup> DON, 100  $\mu$ g kg<sup>-1</sup> ZEN, 50  $\mu$ g kg<sup>-1</sup> OTA, 100  $\mu$ g kg<sup>-1</sup> FB1, 100  $\mu$ g kg<sup>-1</sup> T-2 and 40  $\mu$ g kg<sup>-1</sup> of ergot alkaloid mix, neither biogas production nor its quality were affected <sup>[32]</sup>. In the same study, the semi-continuous AD of contaminated corn (4413  $\mu$ g kg<sup>-1</sup> DON, 729  $\mu$ g kg<sup>-1</sup> 3-ADON + 15-ADON, 14  $\mu$ g kg<sup>-1</sup> AOH, 28  $\mu$ g kg<sup>-1</sup> T-2, 1052  $\mu$ g kg<sup>-1</sup> ZEN, 170  $\mu$ g kg<sup>-1</sup> FB1 + FB2 and >80  $\mu$ g kg<sup>-1</sup> ENNB) using 25 days of HRT demonstrated a stable and productive process, even in the face of a continuous feeding of heavily contaminated material <sup>[32]</sup>. Ferrara et al. <sup>[30]</sup> observed that fumosins' contamination of silage (241.5–13,874  $\mu$ g kg<sup>-1</sup> FB1, 86.5–3877  $\mu$ g kg<sup>-1</sup> FB2, 42.5–3591  $\mu$ g kg<sup>-1</sup> FB3) did not hamper the methane production in batch tests.

These results were in contrast with the findings described by Tacconi et al. <sup>[34]</sup>, who studied the effect of AFB1 on a semi-continuous anaerobic digestion process feed with pig slurry and corn grain. They tested the effects of the increasing AFB1 concentration on AD, operating with 15 days of HRT and high mycotoxin concentrations (25, 50, and 100 µg kg mixture<sup>-1</sup> wet weight). The daily addition of AFB1 concentration higher than 25 µg kg<sup>-1</sup> caused the inhibition of methanogenic bacteria, leading to volatile fatty acids' accumulation, pH decrease and AD failure. Probably, the short HRT and the high organic loading rate used in the experiments could have affected the AD, making the process more susceptible to inhibition mechanisms.

Although most of the literature concerning the AD of mycotoxin-contaminated matrices is regarding corn and silage, contaminated wheat products were also studied in AD <sup>[31][33]</sup>. Seven naturally contaminated flour samples (DON = 0, 1976, 4586 and 10,470  $\mu$ g kg<sup>-1</sup>) or artificially spiked commercial flour (DON = 0, 8000 and 80,000  $\mu$ g kg<sup>-1</sup>), were digested in batch tests. The biogas potential of the wheat flours ranged from 667.2 to 742.8 Nm<sup>3</sup> ton<sup>-1</sup>, demonstrating no significant effect of the DON concentration on the biogas volume and quality produced <sup>[33]</sup>. More recently, Soldano et al. <sup>[31]</sup> investigated the AD of different milling products of durum wheat (whole wheat flour, bran fractions, semolina, and fine middling) contaminated with DON and T-2 + HT-2 toxins. No significant correlations were found between the potential biomethane production and mycotoxins' initial concentrations, independently from the milling fraction.

All the reviewed literature indicates the potential for energy recovery from contaminated feedstock through AD. In fact, it appears clear that biogas production and process stability are not affected by even heavy contaminated

feedstock. The possible long-term inhibition of AD could be easily avoided by interchanging contaminated and noncontaminated feedstock. This strategy could be feasible, considering that mycotoxins' contamination is a seasonal issue.

Concerning nutrients' recovery through agricultural reuse of the digestates obtained from AD of the mycotoxincontaminated feedstock, further assessments are needed. Besides a high fertilizer potential related to the high content of ammonium-N, P and K, the agricultural reuse of digestate faces agronomic and environmental issues (e.g., residual phytotoxicity, high salinity, mycotoxin residues) <sup>[34]</sup>.

#### 2.2. Mycotoxins' Fate during AD

Fate of mycotoxins throughout the AD process is a major concern for the feasible recovery of energy and nutrients from contaminated products. Indeed, digestate utilization depends on the complete removal of mycotoxins, to obtain a safe product that is spreadable on agricultural soil.

Many authors have studied the microbial degradation of mycotoxins <sup>[35][36]</sup> and different bacteria commonly found in AD microflora demonstrated the ability to degrade mycotoxins (i.e., *Pseudomonas* spp. and *Bacillus* spp.) <sup>[37][38]</sup>. In addition, AFB1 has been reported to bind to the bacterial surface of several *Lactobacillus* strains by hydrophobic interactions <sup>[39]</sup>.

The literature agrees that AD can effectively remove mycotoxins from corn- and wheat-contaminated products <sup>[28]</sup> <sup>[29][30][31][32][33]</sup> (**Table 2**). Mycotoxin removal depended mostly on the type of mycotoxin and the operational conditions of AD, whereas the matrix seems to be irrelevant of the removal processes. Overall, the synergic effect of microbial activity, temperature, pH, binding to cell walls, and mineralization is considered responsible for mycotoxin removal during AD.

Some classes of mycotoxins were removed easier during AD with respect to the others. For instance, DON, T-2, and HT-2 demonstrated an average removal higher than 90% in the batch and CSTR mesophilic AD of corn and wheat products <sup>[31][32][33]</sup>. Other mycotoxins demonstrated a lower removal in anaerobic conditions (i.e., the ergot alkaloid mix, and fumosins' concentrations decreased by about 60% and 20–60% in the batch of mesophilic AD, respectively) <sup>[30][32]</sup>. AFB1 was removed with a moderate efficiency during AD, demonstrating a removal range of about 55–90% <sup>[28][30][33]</sup>. Tacconi et al. <sup>[34]</sup> is the only study that reports the AFB1 accumulation during CSTR AD, and it was probably related to the daily addition of heavy contaminated feedstock to the digester combined with a short HRT and a high OLR.

The temperature regime seems to affect mycotoxin removal, whereas batch and CSTR processes did not differ in contaminants' degradation efficiency. De Gelder et al. <sup>[32]</sup> observed significant differences in mycotoxins' removal between the mesophilic and thermophilic batch of AD, with the thermophilic process being more efficient than the mesophilic one. Higher thermophilic degradation has previously been described by other authors for the degradation of several emerging organic pollutants (i.e., polycyclic aromatic hydrocarbons, di-2-(ethyl-hexyl)-phtalate, estradiol, endocrine disrupting compounds, and non-steroidal anti-inflammatory drugs) <sup>[40][41]</sup>.

No evidence of the feedstock composition influence on mycotoxin removal was found in literature, and this is in accordance with Sertillanges et al. <sup>[20]</sup>, who did not report the global influence of the substrate type on organic micropollutants' degradation during AD.

Nowadays, the literature does not detail whether mycotoxins' removal during AD wis due to complete mineralization, binding to cells' walls or transformation to other compounds. Tacconi et al. <sup>[34]</sup> detected the AFB2 during the CSTR digestion of AFB1-contaminated corn grain and they explained it through the acid-catalyzed water addition to the vinylene group of the dihydrofuran moiety of AFB1. This represents a major issue that should be addressed, since mycotoxins should be mineralized or transformed into non-toxic compounds to ensure a safe reuse of digestate in agriculture. The biotransformation of emerging organic contaminants in homologous compounds was also addressed by Zhang et al. <sup>[24]</sup>, for steroid hormones such as androgens, progestogens, and glucocorticoids They demonstrated the negative removal (accumulation) of hormones during biological treatments of contaminated manure due to the bioconversion from hormones' conjugate forms or to the transformation of one hormone into another.

## 3. Composting of Contaminated Products

Differently from AD, the composting of mycotoxin-contaminated matrices is still almost unexplored. Only a few papers reporting the effects of mycotoxins on composting and compost quality can be found in the literature, highlighting the need for further investigation regarding this treatment. **Table 3** presents the main findings about the composting of mycotoxin-contaminated products.

Mycotoxin	Initial Contamination (µg kg <sup>-1</sup> )	Organic Substrate	Composting Process	Peak Temperature (°C)	Average Mycotoxin Removal	References
		Corn grain and pig slurry	Pilot scale,	75.5	85.7%	
AFB1	100	Corn grain and organic fraction of municipal solid wastes	passive aerated, static composting	74.8	97.3%	[ <u>42</u> ]
AFB1 + AFB2 + AFG1 + AFG2	195.4 (AFB1) + 22.2 (AFB2) + 2.9 (AFG1) + 1.2 (AFG2)	Peanut meal	Laboratory scale, actively aerated, continuously mixed composting	36.4	58.6% (AFB1) 54.5% (AFB2) 96.6% (AFG1)	[43]

Table 3. Composting of mycotoxin-contaminated products: process characteristics and mycotoxins' fate.

Mycotoxin	Initial Contamination (µg kg <sup>-1</sup> )	Organic Substrate	Composting Process	Peak Temperature (°C)	Average Mycotoxin Removal	References
					83.3% (AFG2)	
	2955 (total AF)	Peanut seeds, peanut shells, peanut leaves, and cowpea pods	Pilot scale, actively aerated, 3- times a week mixed composting	n.a.	77%	
ΟΤΑ	0.37–1.66	Coffee pulp and husks + bulking material	Real scale, passive aerated, monthly mixed composting	n.a.	400–600% *	[44]

only in the last few years <sup>[42][43]</sup>. These studies evaluated the effect of mycotoxins on the composting evolution, compost quality, and mycotoxins' fate (**Table 3**). The temperature behavior during composting is an adequate realtime indicator for the optimal conditions for supporting the mitcrobial activity and the organic matter degradation. Moreover, a minimum temperature of 55 °C must be maintained for three days during the thermophilic phase to obtain biomass hygenization <sup>[15]</sup>. An effective composting process should also produce mature and stable compost, and it can be assessed through C/N ratio determination, phytotoxicity assays, and a water-soluble organic matter analysis <sup>[15][45][46][47]</sup>. Finally, the absence or low level of toxic compounds (e.g., heavy metals) should be attained after composting to achieve the safe recycling of plant nutrients.

Akoto et al. <sup>[43]</sup> reported the first assessment of composting for peanut meal decontamination from aflatoxins. They carried out both laboratory scale and pilot scale experiments using peanut by-products contaminated with AFB1, AFB2, AFG1, and AFG2. Temperature profiles demonstrated a regular behavior, indicating that aflatoxins did not produce toxic effects on thermophilic microflora. Compost obtained from the pilot scale experiment demonstrated acceptable contents of nitrogen, phosphorous, potassium and micronutrients, as well as a high maturation (C/N ratio was about 4.5) and low content of heavy metals.

Results from Akoto et al. <sup>[43]</sup> were confirmed in a later study where AFB1 contaminated corn grain (100 µg kg<sup>-1</sup> AFB1, wet weight) was co-composted using two different co-substrates (pig slurry and organic fraction of municipal solid wastes) <sup>[42]</sup>. AFB1 did not affect the temperature profile during the active phase of composting, and a high temperature were reached (75.5 °C and 74.8 °C for the pig slurry and organic fraction of municipal solid waste mixtures, respectively). AFB1 did not affect maturation and stabilization processes during the composting, and the final products were characterized by an optimal C/N ratio (about 10), absence of phytotoxicity (germination index was higher than 100%, probably due to high nutrients' concentration and the presence of phytohormone-like substances with biostimulant activity) and reduced content of water-soluble organic matter.

Preliminary studies demonstrated that the composting process evolution and compost quality are not affected by mycotoxins' contamination of the feedstock. Similar results were obtained by other authors, who demonstrated that antibiotics, heavy metals, and polycyclic aromatic hydrocarbons contaminations of the composting mixture do not negatively affect the composting process and compost quality <sup>[47][48][49]</sup>. Nevertheless, the actual knowledge is still limited to the investigation of limited classes of mycotoxins (mainly aflatoxins) and small-scale experiments. Although the potential for nutrients' recovery from mycotoxin-contaminated products through composting appears evident, deeper studies are needed to up-scale this treatment.

#### 3.2. Mycotoxins' Fate during Composting

The fate of mycotoxins during the composting of contaminated products was assessed to understand whether aerobic biological treatments can represent a suitable strategy for decontamination.

As already described for AD, a reduction in mycotoxins' concentration during composting is expected, since natural composting microorganisms were reported to efficiently degrade some classes of mycotoxins (e.g., aflatoxins) <sup>[50]</sup>. For instance, fungi (*Armillariella tabescens*) and bacteria (*Pseudomonas putida*) species can degrade AFB1 into less toxic metabolites (aflatoxin D and dihydrodiol-derivates) through two different pathways: (1) modification of the difuran ring and (2) modification of the coumarin structure. Recently, the effective degradation of AFB1 using a thermophilic microbial consortium extracted from compost produced from agricultural wastes was described by Wang et al. <sup>[51]</sup>. They observed a 95% degradation of AFB1, with an optimal temperature of 55–60 °C and an optimal pH of 8–10. Moreover, the thermophilic microbial consortium exhibited the tolerance to high doses of AFB1 (up to 5000  $\mu$ g L<sup>-1</sup>) and extreme heat.

The literature review confirmed that aflatoxins are effectively degraded by composting microorganisms, and aflatoxins' removal is comparable to the one reported for AD (**Table 3**). Akoto et al. <sup>[43]</sup> reported a 58.6, 54.5, 96.6, and 83.3% removal for AFB1, AFB2, AFG1, and AFG2, respectively, after the laboratory scale composting of contaminated peanut meal. When contaminated peanut by-products were composted in a pilot scale composting pile, 77% of removal was observed for the total aflatoxins.

The pilot scale co-composting of AFB1-contaminated corn grain with pig slurry or the organic fraction of municipal solid wastes reduced the AFB1 content from 13.04  $\mu$ g kg<sup>-1</sup> AFB1 and 12.20  $\mu$ g kg<sup>-1</sup> AFB1 to 0.35  $\mu$ g kg<sup>-1</sup> AFB1 and 1.75  $\mu$ g kg<sup>-1</sup> AFB1, respectively <sup>[42]</sup>. The average AFB1 removal was 91.5%, a remarkable result that was probably related to the synergic effects of several decontamination agents (microbial activities, high temperature during the active phase, high ammonium-N concentration, and light irradiation).

Composting has already been reported to be an effective biological treatment for organic contaminants' reduction in organic wastes <sup>[19]</sup>. For instance, Cucina et al. <sup>[47]</sup> reported that the antibiotic daptomycin was degraded during co-composting through a protease-mediated mechanism. Similarly, extracellular enzymes produced by composting microorganisms may hydrolyze mycotoxins, making their mineralization easier.

Differently from aflatoxins, Ochratoxin A (OTA) increased steadily with the progress of the composting process of coffee pulp and husk, alone or in combination, in naturally and artificially contaminated compost <sup>[44]</sup>. Authors explained the increase in OTA content with the presence of OTA-producing fungi such as *Aspergillus* spp. section *Nigri*. Since this represents the only report on the OTA fate during composting, further studies are needed to assess whether composting can reduce OTA contamination.

As reported for AD, the literature has not yet evaluated whether mycotoxins removal at the end of composting is due to mineralization or other mechanisms (e.g., binding to cell walls or transformation to other compounds). This aspect should be evaluated in depth to ensure that the compost obtained from mycotoxin-contaminated products is safe and free from toxic compounds.

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