Received: 13 July 2023, Accepted: 03 September 2023

Journal of Advanced Veterinary Research
(2023) Volume 13, Issue 8, 1726-1729

Review Article

Mould Contamination of Fish and Fish Products with a Special Reference to its Public Health Significance: A Review

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Abstract

Fish and fish products represent major contributors to supply humans with part of their needs of the essential amino acids, vitamins, minerals, omega -3-fatty acids, and other needed micronutrients. However, fish and fish products are highly perishable foods that can easily spoiled and decomposed, possibly because of its cross contamination from the surrounding environment. Fish attracts a vast array of microorganisms, of these; mould and yeast represent a major sector of these microbiota, which by turn van lead to rapid decomposition of fish or even produce several toxicological implications if ingested. In this review, we highlighted the available literature about mould contamination of fish and fish products with a special reference to its public health significance.

KEYWORDS
Fish, Fish products, Mould, Mycotoxins, Public health

Introduction

Aquaculture is one of the fastest-growing food businesses, accounting for nearly 50% of global fish consumption in 2011 (FAO, 2013). In low- and middle-income nations, small-scale farmers produce the majority of farmed fish (>70%) in fresh water (Hastein *et al*., 2006). While fish is a good source of low-fat, protein-rich foods high in omega-3 and omega-6 fatty acids, which protect against adverse health consequences such as coronary heart disease and stroke (Morshdy *et al*., 2019, 2021). Egypt is one of the top ten producers internationally and the major producer of aquaculture in Africa, with 1.5 million tons produced in 2015 (Eltholth *et al*., 2015). There is also rising concern about probable chemical and microbiological foodborne contamination (Morshdy *et al*., 2022). As a result of these concerns, demand for farmed fish may fall (Smallwood and Blaylock, 1991). This may have a detrimental influence on fish farmers, as well as a decrease in the consumption and use of animal-derived goods.

Mould contamination of fish and fish products reflects the sanitary status and hygienic methods used during fish handling, starting from catching, evisceration, transportation, and chilling or freezing. Mould spores can be found in a variety of environments, including animal waste, air, water, and soil. Mould contamination of fish and fish products can also be caused by operators' hands, equipment, and utensils. One element that promotes mould formation is the occurrence of fluctuating chilling and freezing temperatures. Mould contamination of a certain food matrix, particularly of aflatoxigenic strains, has an impact on both the quality and safety of that food (Darwish *et al*., 2016). There is little information available about the mycological status of the fish and fish products. In this review, we would like to highlight studies focusing on the fungal status of the fish and fish products with a special reference to the public health significance of the most frequently recovered mold species.

Prevalence of different mould genera in fish and fish products

In Nigeria, Twenty of the 33 moulds identified from 20 samples of wood-smoked *Chlamydoselachus anguineus* (shark-fish) produced compounds harmful to viable Hubbard Golden Comet (Niger chick) eggs. Mould isolates from *Aspergillus* and *Pencillium* were the most common. Eurotium, Fusarium, and *Cladosporium* species were also discovered as toxigenic moulds. The ability of the isolates to produce protease varied between genera and between isolates of the same species. The presence and growth of these moulds on smoked fish indicates the potential health risk connected with eating mouldy dry fish (Essien *et al*., 2005). Besides, Adebayo-Tayo *et al*. (2008) aimed to estimate the mycoflora and aflatoxin contamination of smoked dried fishes of Stock (*Gadus morhua*), Skip jack tuna (*Katsuworus pelamis*), Croaker (Pseudotolithus typhus), Sting ray (Dasyatis margarita), Cat (*Arius hendeloti*), Bonga (*Ethalmosa fimbriota*), Ribban fish (Triuchurius trichurius), Stark (*Carchanas faunis*), Thread fin (Pentanemis qumquarius), Sole (*Cynoglossus browni*), Spade (*Drepane africana*) in Uyo, eastern Nigeria. Fifty-five smoke-dried fish samples sold at three distinct marketplaces in Uyo town, Itam and Akpan Adem, in Uyo, Akwa Ibom state, Nigeria, were significantly infected with mould. Twelve distinct fungus were discovered to be related with smoked dried fish samples offered in three separate markets. *Aspergillus flavus* and *Aspergillus tereus* were the fungus found in the area. *Absidia* sp., *Rhizopus* sp., *Aspergillus fumigatus*, *Mu-*

ISSN: 2090-6277/2090-6269/

cor sp. Among the isolated fungus, *Cladosporium* sp., *Penicillium Italiculum*, *Penicillium viridatum*, *Candida tropicalis*, *Fusarium moniliformis, Aspergillus flavus*, and *A. tereus* had the highest rate of occurrence. Aflatoxin B and G were discovered to be related with the samples. The amounts of Aflatoxin B and G in the sample ranged from 1.5 to 8.1 g/kg and 1 1 g to 1.8-4.5 g/kg, respectively. The fungal counts ranged from 3.0x10 to 4.8x10 cfu/g. The moisture content and pH ranged from 22.7 to 27.6% and 3.0 to 6.0, respectively. In addition, Junaid *et al*. (2010) isolated and identified the fungus linked to stockfish pollution in Jos Metropolis. A total of 100 stockfish samples were acquired at random from four marketplaces in Jos, Plateau State, Nigeria: Terminus, Kwararafa, Katako, and Gada biu. Using normal techniques, the stockfish samples were tested for fungal infection and moisture content. Fungi infected all of the stockfish samples. Seven distinct fungus were discovered to be linked to stockfish samples sold in four separate markets. *Mucor* Spp, *Aspergillus flavus*, *Trichophyton verrucosum*, *Aspergillus* niger, *Aspergillus fumigatus*, Penicillin Spp, and *Rhizopus* Spp were the related fungi. *Mucor* Spp. was found to have the greatest incidence of occurrence among the isolated fungus. The moisture content ranged from 6 to 27%. Moreover, Olajuyigbe *et al*. (2014) collected forty fisheries items (27 fin fish, 4 shell fish, and 9 fish feed samples) from Lagos markets and tested for the presence of aflatoxigenic moulds and aflatoxins in order to assess the quality of these products. The dilution plating technique was used for mycological study, and an Enzyme-Linked Immunosorbent Assay method was used for aflatoxin measurement. The only *Aspergillus* species were *A. flavus* and *A. tamarii*. *Flavi* species were recovered from all fish feed samples, 50% of shellfish samples, and 37% of fin fish samples. In each type of fisheries product, *A. flavus* was more common than *A. tamarii*. In all categories of fisheries products, the incidence of non aflatoxigenic *A. flavus* isolates was higher than that of aflatoxigenic *A. flavus* isolates. Aflatoxin concentrations ranged from 1.05-25.00 g/kg in all fish and fish feed samples. Aflatoxin levels in fish feed samples were substantially higher than in fish samples. Total aflatoxin levels in fish diet surpassed 10 g/kg in 66.67% and 22.22% of samples, respectively. In light of mycotoxin contamination, smoked-dried fin and shellfish may constitute a safe food for human consumption. Furthermore, efforts should be increased to reduce aflatoxin levels in fish feed formula.

In Benin, Anihouvi *et al*. (2019) determined the microbiological status of smoked fish (SF) and smoked-dried fish (SDF) processed in Benin, as well as the contamination variables related with these products. A total of 66 fresh and processed fish samples were obtained at random from various processing plants and markets for microbial characterization using conventional procedures. The density of aerobic mesophilic bacteria (AMB) ranged from 2.9 to 9.5 Log_{10} CFU/g. 63.9%, 27.8%, 55.6%, 58.3%, 61.1%, and 77.8% of samples included *Enterobacteriaceae*, *Escherichia coli*, *Bacillus cereus*, *Clostridium perfringens*, yeasts, and moulds, respectively, whereas no *Salmonella* spp., *Listeria monocytogenes*, or *Staphylococcus aureus* were detected. The bulk of SF samples (66.7%) and 22.2% of SDF samples did not meet the permissible limit of 7.0 Log_{10} CFU/g recommended by the Health Protection Agency for AMB. Whereas 50% of SF and 5.6% of SDF samples had Enterobacteriaceae levels that above the acceptable range of 4.0 Log_{10} CFU/g. Similarly, 38.9% of SF samples tested positive for E. coli. Microbiological hazard analysis of practices enabled the identification of sensitive phases where hygiene measures should be emphasized for better quality control.

In Korea, Park and Ha (2015) tested how cold oxygen plasma (COP) affected the decreases of *Penicillium citrinum* and *Cladosporium* cladosporioides on the surface of dried filefish fillets (*Stephanolepis cirrhifer*). The numbers decreased significantly (P 0.05) as the treatment time (3-20 min) of COP on the fillets increased. However, no significant (P > 0.05) variations in counts between 3 and 5 minutes of COP were noticed. The average reduction in C. cladosporioides and P. citrinum counts generated by 3-20 minutes of COP was 0.91 and 1.04 log_{10} CFU g^1, respectively. On fillets treated with COP for more than 10 minutes, a decrease of >1-log_{10} CFU g^1 was observed. The Weibull model's decimal reduction time (dR) was 9.32 and 7.42 minutes, respectively for *C. cladosporioides* and *P. citrinum*, respectively. The fillets exposed to COP for 20 minutes had higher levels of thiobarbituric acid reactive substance (TBARS) and lower overall sensory acceptability. However, fillets treated with 10 minutes of COP had acceptable TBARS and consumer acceptability. As a result, a 10-min COP could be beneficial in lowering >90% and inactivating the mould without affecting the physicochemical and sensory properties of the fillet.

In Indonesia, Indriati *et al*. (2017) investigated the presence of aflatoxin B1 in commercial dried salted fish and other relevant data. Based on the salt concentration, 150 samples were divided into three groups. Dry anchovies (*Stolephorus* sp.) and commerson's anchovy (*Stolephorus commersonii*) had low salt content (0-5%); medan anchovy (*Stolephorus bataviensis*) and whipfin silverbiddy (*Gerres filamentosus*) had moderate salt content (6-10%); and snakehead fish (*Channa striata*) had high salt content (>10%). *Aspergillus flavus*, Aflatoxin B1, salt concentration, moisture content, pH, water activity, and total mould count were all assessed in samples gathered from various sellers on Java Island. The dried salted fish were contaminated with *A. flavus* at 25.2-32.2ºC, 65-84% humidity, 17-50% moisture content, and 0.25-19.88% salt content and 0.73-0.86 aw. The incidence of *A. flavus* in dried salted fish was 9.33% (14/150), and aflatoxin B1 was 8% (12/150), with detectable values ranging from 10.71 to 33.6 ppb.

In Egypt, Gouda (2015) mentioned that information about fungi associated with food and feeds is important in assessing risk of mycotoxin contamination. Mould contamination not only causes deterioration of food and feeds, but also can adversely affect the health of humans and animals, once they may produce toxic metabolites. Moulds are capable of reducing the nutritional value of feedstuff as well as elaborating several mycotoxins. Mycotoxin-contaminated feed has adverse effects on chicken, fish and animal health and productivity. In point prevalence study feedstuff used for chicken and fish nutrition in Egypt was analyzed for fungal flora and natural incidence of selected my cotoxins. Analyzed fungal flora of chicken and fish feed samples which collected from three different localities i.e. Cairo, Qalubiya and Sharkiya governorates in Egypt yielded 885 fungal isolates. Fish feed samples had a greater percentage of total fungal colonies (about 54.6%) than chicken feed samples (45.4%). From these samples, seven fungal species from four fungal genera were isolated and identified. *Aspergillus* (*A. flavus*, *A. parasiticus, A. niger*, and *A. ochraceous*), *Penicillum, Fusarium*, and *Alternaria* spp. are the genera involved. Six fungal isolates associated with fish feeds were found to produce one or more mycotoxin, namely aflatoxin(s), ochratoxin A (OTA), and fumonisin B1 (FB1). Of these, five fungal isolates of *A. flavus*, one of *A. parasiticus*, were found to produce aflatoxin(s), two isolates of *A. ochraceous*, as well as two isolates of *Penicillum* sp., were found to produce Fumonisin FB1. Besides, Mostafa *et al*. (2019) determined the fungal status of two regularly consumed fish species in Egypt, *Tilapia nilotica* and *Mugil cephalus*. The second trial looked at the antifungal effect of natamycin on *Tilapia nilotica*. During the winter season of 2018, 60 fish samples, comprising *Tilapia nilotica* and *Mugil cephalus* (30 of each), were randomly gathered from several retail markets and stores with varying cleanliness levels in Kafrelsheikh Governorate, Egypt. Fungal contamination was checked on all samples. Mould counts in *Tilapia nilotica* and *Mugil cephalus* samples were 3.6310sup>2/sup> and 1.6510sup>2/sup>, respectively. From two fish species, nine fungal species were isolated. *Tilapia nilotica Mugil cephalus* yielded seven and five species, respectively. *Aspergillus flavus* was the most common fungal species isolated from the two fish. Natamycin demonstrated substantial antifungal activity in a concentration-dependent manner. Thus, efficient hygienic handling and timely cooling of fish can help to reduce fungal contamination. Furthermore, soaking or spraying fish with natamycin solution is an effective technique for lowering the fungal load of raw fish. According to El Bayomi *et al*. (2021) thyme, cinnamon, and lemongrass essential oils exhibit high antifungal activity, which is a viable option for mould decontamination and, as a result, improves the shelf life of fish and meat products.

Public health significance of moulds frequently isolated from foods

Several mould species are frequently isolated and recovered from meat, fish, chicken, and their products. Of these, moulds that belong the *Aspergillus* group are the most recovered. A. niger is one of the most common predominant species in several studies. A. niger causes severe allergic reactions, pulmonary aspergillosis, and produce toxic metabolites such as oxalic acid, kojic acid, malformins (Bennett, 1980). *A. flavus* is also one major *Aspergillus* species that is commonly isolated from different food matrices. *A. flavus* causes Aspergilloma, allergic bronchopulmonary aspergillosis, craniocerebral aspergillosis, and produce toxic metabolites like aflatoxins, aspergillic acid, kojic acid, asperotoxin, and sterigatocystin (Denning *et al*., 2003). A. *fumigatus* also induces Aspergillosis, aspergilloma and allergic reactions, and produces toxic metabolite called gliotoxin (Chakrabarti *et al*., 2002). *A. parasiticus* produces toxic metabolites called aflatoxins, while *A. ochraceous* produces ochratoxin A and citrinin (Darwish *et al*., 2014, 2016). *Penicillium* spp. are also a large group of moulds that can produce several metabolites that cause health hazards including cyclopiazonic acid (organ damage in mammals), meleagrin (mutagenic), mycophenolic acid (immunosuppressive), penitrem A (tremorgenic), roquefortine C (neurotoxic), rugulovasine A (Anti-hypotensive), terrestric acid (cardiotoxic) (Pitt and Hocking, 2009). *Cladosporium* spp. causes allergic reactions (Schoch *et al*., 2006). *Mucor* spp. causes allergic reactions and pneumonia (Patriarca *et al*., 2014). While Fusraium spp., causes fusariosis, and peritoneal dialysis (Tupaki-Sreepurna and Kindo, 2018).

Aflatoxins (AFTs) are secondary metabolites generated by *A. flavus* and *A. parasiticus*, among others (Alcaide-Molina *et al*., 2009). AFT contamination of fish and fish products may begin during the aquaculture life by intake of contaminated feed and water or as metabolites caused by the growth of specific fungus on the fish flesh substrate (El-Ghareeb *et al*., 2013). Then, AFTs enter the human body through contaminated foods, causing a variety of health problems (Darwish *et al*., 2014). AFTs are known to be mutagenic, carcinogenic, particularly for hepatocellular carcinoma, and immunosuppressive (Aljazzar *et al*., 2021, 2023; Darwish *et al*., 2022).

Conclusion

The current review threw the light on the fungal status of the fish and fish products, which plays an important role in food security worldwide, particularly for the protein of animal origin. Fish and fish products can be contaminated with mould of different species which might produce several adverse health effects. Therefore, it is necessary to adopt restrict food safety measures to ensure continuous monitoring and checking of the fungal quality of fish and fish products such as dried, salted and smoked fish.

Conflict of interest

The authors declare that they have no conflict of interest.

References

Adebayo-Tayo, B.C., Onilude, A.A., Patrick, U.G., 2008. Mycofloral of smoke-dried fishes sold in Uyo, Eastern Nigeria. World J. Agricul. Sci. 4, 346-350.

Alcaide-Molina, M., Ruiz-Jiménez, J., Mata-Granados, J.M., Luque de Castro, M.D., 2009. High through-put aflatoxin determination in plant material by automated solid phase extraction on-line coupled to laser-induced fluorescence screening and determination by liquid chromatography–triple quadrupole mass spectrometry. J. Chrom. A 1216, 1115–1125.

Aljazzar, A., El-Ghareeb, W.R., Darwish, W.S., Abdel-Raheem, S.M., Ibrahim, A.M., 2021. Content of total aflatoxin, lead, and cadmium in the bovine meat and edible offal: study of their human dietary intake, health risk assessment, and molecular biomarkers. Environ. Sci. Pollut. Res. 28, 61225-61234.

Aljazzar, A., El-Ghareeb, W.R., Darwish, W.S., Abdel-Raheem, S.M., Ibrahim, A.M., Hegazy, E.E., Mohamed, E.A., 2023. Effects of aflatoxin B1 on human breast cancer (MCF-7) cells: cytotoxicity, oxidative damage, metabolic, and immune-modulatory transcriptomic changes. Environ. Sci. Pollut. Res. 30, 13132-13140.

Anihouvi, D.G.H., Kpoclou, Y.E., Abdel Massih, M., Iko Afe, O.H., Assogba, M.F., Covo, M., Scippo, M.L., Hounhouigan, D.J., Anihouvi, V., Mahillon, J., 2019. Microbiological characteristics of smoked and smoked–dried fish processed in Benin. Food Sci. Nutr. 7, 1821-1827.

Bennett, J.E., 1980. Aspergillosis, pp. 742-744. In K.J.Chakrabarti, A., Sethi, S., Raman, D. S., Behera, D., 2002. Eight year study of allergic bronchopulmonary aspergillosis in an Indian teaching hospital. Mycoses 45, 295–299.

Darwish, W., Bayomi, R. M., El-Moaty, A. M., Gad, T. M., 2016. Mould contamination and aflatoxin residues in frozen chicken meat-cuts and giblets. Jpn. J. Vet. Res. 64, S167–S171.

Darwish, W. S., Ikenaka, Y., Nakayama, S. M., Ishizuka, M., 2014. An overview on mycotoxin contamination of foods in Africa. J. Vet. Med. Sci. 76, 789–797.

Darwish, W.S., El-Ghareeb, W.R., Alsayeqh, A.F., Morshdy, A.E.M., 2022. Foodborne intoxications and toxicoinfections in the Middle East. In Food Safety in the Middle East. Academic Press. pp. 109-141.

Denning, D. W., Riniotis, K., Dobrashian, R., Sambatakou, H., 2003. Chronic cavitary and fibrosing pulmonary and pleural aspergillosis: case series, proposed nomenclature and review. Clin. Infect. Dis. 37, S265–S280.

El Bayomi, R.M., Hebishy, R.M., Darwish, W.S., El-Atabany, A.I.M., Mahmoud, A.F.A., 2021. Mould contamination of some meat products with reference to decontamination trials of *Aspergillus flavus* using essential oils. Slov. Vet. Res. 58, 363-372.

El-Ghareeb, W. R., Darwish, W. S., Tharwat, A. E., El-Desoky, K. I., Hussein, M. A., 2013. Aflatoxin and ochratoxin A residues in some meat additives. Life Sci. J. 10, 3411–3417.

Eltholth, M., Fornace, K., Grace, D., Rushton, J., Häsler, B., 2015. Characterisation of production, marketing and consumption patterns of farmed tilapia in the Nile Delta of Egypt. Food Policy 51, 131-143.

Essien, J.P., Ekpo, M.A. and Brooks, A.A., 2005. Mycotoxigenic and proteolytic potential of moulds associated with smoked shark fish (*Chlamydoselachus anguincus*). J. Appl. Sci. Environ. Manag. 9, 53-57.

FAO, 2013. FAO Fisheries and Aquaculture Department has published the Global Aquaculture Production Statistics for the year 2011. 2013 ed.: ftp.fao.org/FI/news/GlobalAquacultureProductionStatistics2011.pdf

Gouda, M.M., 2015. Mycoflora and mycotoxin contaminated chicken and fish feeds. Sciences 5, 1044-1054.

Hastein, T., Hjeltnes, B., Lillehaug, A., Utne Skare, J., Berntssen, M., Lundebye, A.K., 2006. Food safety hazards that occur during the production stage: challenges for fish farming and the fishing industry. Rev. Sci. Tech. 25, 607-625.

Indriati, N., Hermana, I., Hidayah, I., Rahayu, E.S., 2017. Prevalence of aflatoxin B1 in commercial dried fish from some regions of Java.

Squalen Bull. Marine Fish. Postharvest Biotechnol. 12, 107-115.
Junaid, S.A., Olarubofin, F., Olabode, A.O., 2010. Mycotic contamination of stockfish sold in Jos, Nigeria. J. Yeast Fungal Res. 1, 136-141.
Morshdy, A.E.M., Darwish, W.S., Daoud, J.R.M., Sebak, M.A.M., 2019. Estimation of metal residues in *Oreochromis niloticus* and *Mugil cephalus* intended for human consumption in Egypt: a health risk assessment study with some reduction trials. J. Cons. Protect. Food Saf. 14, 81-91.
Morshdy, A.E., Darwish, W.S., Hussein, M.A., Mohamed, M.A., Hussein, M.M., 2021. Lead and cadmium content in Nile tilapia (*Oreochromis niloticus*) from Egypt: a study for their molecular biomarkers. Sci. Afr. 12, 1-8.
Morshdy, A. E. M., Hussein, M. A., Mohamed, M. A. A., Hamed, E., El-Murr, A. E., Darwish, W. S., 2022. Tetracycline residues in tilapia and catfish tissue and the effect of different cooking methods on oxytetracycline and doxycycline residues. J. Cons. Protect. Food Saf. 17, 387-393.
Mostafa, N.Y., Kirrella, G.A., Aideia, H.A., Abo Shaisha, J.M., 2019. Assesstment of mould contamination of *Tilapia nilotica* and *Mugil cephalus* fish and trials to reduce using natamycin. Slov. Vet. Res. 56, 515-522.
Olajuyigbe, O.O., Akande, G.R., Ezekiel, C.N., Ezekiel, M.O., 2014. Aflatoxigenic moulds and aflatoxin contamination of retailed fishery products in Lagos markets. Mycotoxicology 1, 57-63.
Park, S.Y., Ha, S.D., 2015. Application of cold oxygen plasma for the reduction of *Cladosporium cladosporioides* and *Penicillium citrinum* on the surface of dried filefish (S tephanolepis cirrhifer) fillets. Inter. J. Food Sci. Technol. 50, 966-973.
Patriarca, A., Vaamonde, G., Pinto, V.F., 2014. Alternaria. In Encyclopedia of food Microbiology. 2nd edition, Academic Press. Pages 54–60.
Pitt, J.I., Hocking, A.D., 2009. Fungi and Food Spoilage, 3rd Ed. Published by Blackie Academic and Professional Academic Press New York, London.
Schoch, C. L., Shoemaker, R. A., Seifert, K. A., Hambleton, S., Spatafora, J. W., Crous, P. W., 2006, A multigene phylogeny of the Dothideomycetes using four nuclear loci. Mycologia 98, 1041-1052.
Smallwood, D., Blaylock, J., 1990. Food Safety and the Demand for Food: Methodological Models and Application. In Economics of Food Safety Workshop," Alexandria VA.
Tupaki-Sreepurna, A., Kindo, A.J., 2018. Fusarium: The versatile pathogen. Indian J. Med. Microbiol. 36, 8-17.