



## Fungal contamination in *Colossoma macropomum* (Cuvier, 1816) from fish farms on Maranhão island

Contaminação fúngica em *Colossoma macropomum* (Cuvier, 1816) oriundos de cultivos da ilha do Maranhão

Douglainny Barros do Nascimento<sup>1</sup>, Franciely Assunção Matão<sup>2</sup>, Greiciene dos Santos de Jesus<sup>3</sup>,  
Danilo Cutrim Bezerra<sup>2</sup>, Amanda Mara Teles<sup>1</sup>, Nancyleni Pinto Chaves Bezerra<sup>1, 2, 3\*</sup>

**ABSTRACT:** The aim of the present study is to assess fungal contamination in *Colossoma macropomum* from fish farms on Maranhão Island. In order to do so, 40 male and female *C. macropomum* (tambaqui) specimens at the fattening phase were collected from four fish farms for further assessment. Muscle fragments were removed and it was followed by enumerating and identifying macroscopic morphology, colonial features and microscopic morphology of filamentous fungi in laboratory environment. Enumeration showed filamentous fungi in 67.5% (n=27/40) of the assessed specimens; fungal populations ranged from  $1.0 \times 10^1$  to  $4.52 \times 10^4$  CFU/gram. Genera *Aspergillus* sp., *Penicillium* sp., *Paecylomeces* sp. and *Rhizopus* sp. were identified through microscopic morphology associated with macroscopic morphology. The applied identification allowed identifying two *Aspergillus* species, namely: *A. niger* and *A. terreus*; this last species presented mycotoxigenic potential. According to the results, the analyzed *C. macropomum* specimens recorded high fungal contamination after two potentially mycotoxigenic genera were identified. Fish is a nutritionally rich seafood type whose intrinsic features, in addition to the conditions observed in the sampled fish farms, which were favorable to fungi survival and multiplication, favor its deterioration. These fungi require more effective control and monitoring, including feed storage in the assessed farms. The goal of all these precautions is to rule out the likely occurrence of toxin-producing fungi capable of compromising the quality of this animal product, which has impact on production and poses risks inherent to consumers' health.

**Keywords:** filamentous fungi; mycotoxins; fish; food security.

**RESUMO:** Objetivou-se verificar a ocorrência de contaminação fúngica em *Colossoma macropomum* oriundos de cultivos da Ilha do Maranhão. Para isso, foram avaliados 40 exemplares de *C. macropomum* (tambaquis), entre machos e fêmeas, em fase de engorda, oriundos de quatro pisciculturas. Em ambiente laboratorial, foram removidos fragmentos musculares seguido da enumeração e identificação de fungos filamentosos (morfologia macroscópica, características coloniais e morfologia microscópica). Na enumeração, constataram-se fungos filamentosos em 67,5% (n= 27/40) dos exemplares avaliados, com populações fúngicas que variaram de  $1.0 \times 10^1$  a  $4,52 \times 10^4$  UFC/grama. Por meio da morfologia microscópica, associada à macroscópica, foram identificados os gêneros *Aspergillus* sp., *Penicillium* sp., *Paecylomeces* sp. e *Rhizopus* sp. Com as chaves de identificação utilizadas, duas espécies de *Aspergillus* foram identificadas, *A. niger* e *A. terreus*, a última espécie com potencial micotoxigênico. Com base nos resultados obtidos, concluiu-se que os exemplares de *C. macropomum* analisados apresentaram elevada contaminação fúngica, com a identificação de dois gêneros potencialmente micotoxigênicos. Os peixes, por constituírem um tipo de pescado rico nutricionalmente, dotados de características intrínsecas que favorecem sua deterioração, acrescidos às condições nas pisciculturas amostradas favoráveis à sobrevivência e multiplicação de fungos, requerem controle e fiscalização mais efetivos, que perpassam pelo armazenamento das rações ofertadas nas criações. Todos esses cuidados visam eliminar a possibilidade de ocorrência de fungos produtores de toxinas, que podem comprometer a qualidade desse produto de origem animal, impactando a produção e com riscos inerentes à saúde dos consumidores.

**Palavras-chave:** fungos filamentosos; micotoxinas; peixes; segurança alimentar.

## INTRODUCTION

Filamentous fungi (mold or mildew) are eukaryotic, heterotrophic and multicellular organisms distributed in all environment types, besides being economically important for medicine, phytopathology, and the food and pharmaceutical industry; and environmentally important decomposers. However, they can also contaminate food and spoil it, and it leads to decreased nutritional value and altered organoleptic

features. Fungi can become a public health issue for causing diseases in animals and humans (Vecchia; Castilhos-Fortes, 2007; Arruda; Beretta, 2019).

The secondary metabolism of some fungi can produce harmful mycotoxins (Franco; Landgranf, 2008) such as aflatoxin, ochratoxin A, zearalenone, patulin, fumonisin, trichothecene, sterigmatocystin, mainly citrinin (Rodriguez-Amaya; Sabino, 2002; Arruda; Beretta, 2019). Researchers have reported more than 100 mycotoxigenic fungal species that can produce more

<sup>1</sup> Professional Graduate Program in Animal Health Defense, State University of Maranhão, São Luís/MA, Brazil.

<sup>2</sup> Graduate Program in Animal Production, State University of Maranhão, São Luís/MA, Brazil.

<sup>3</sup> Graduate Program in Animal Science, State University of Maranhão, São Luís/MA, Brazil.

\*Corresponding author: nancylenichaves@hotmail.com.

Received: 10/15/2024.

Accepted: 01/10/2026.

than 400 different mycotoxins types, altogether; and more than 250 of them already have their chemical structures defined (Paterson; Venâncio; Lima, 2004; Pereira *et al.*, 2005; Rosa *et al.*, 2006; Simas; Botura; Correa, 2007).

The main fungi producing mycotoxins belong to genera *Aspergillus*, *Fusarium*, *Penicillium*, *Alternaria*, *Rhizoctonia* and *Stachybotrys*. The first three genera stand out as the most important ones for food and feed because they are the most frequent ones, besides being reported as the main producers of the aforementioned secondary metabolites. However, their biological impact on human and animal health, and on the environment, remain somehow unclearly (Knasmüller *et al.*, 2004). Mycotoxigenic fungi can be found in different food types consumed by humans and animals (Batista; Freitas, 2000; Pereira *et al.*, 2005; Hermanns *et al.*, 2006; Chiotta *et al.*, 2009; Cardoso Filho *et al.*, 2013).

Agricultural and food products contaminated by both toxigenic fungi and toxins have negative economic impact; therefore, this matter has gotten closer attention in recent decades, mainly in developing countries like Brazil. Appropriate harvesting, post-harvest and processing techniques must be implemented, as well as adequate storage conditions, including temperature and humidity, in order to prevent these microorganisms from developing in food (Tacon; Metian, 2008; Griessler; Encarnação, 2009; Filho; Campolina; Dias, 2013). Although mycotoxigenic fungi can be found in different substrates, the ones showing high carbohydrate content are more prone to contamination (Vecchia; Castilhos-Fortes, 2007).

Several ingredients are used in fish farming for feed formulation as the way to meet fish nutritional needs at different production-cycle phases. These feed types are ideal substrates for fungal growth and for the production of their metabolites, if the favorable conditions are provided (Dantigny; Guilmar; Bensoussan, 2005; Cardoso Filho *et al.*, 2013).

Cardoso Filho *et al.* (2013) assessed 36 fish-feed samples to set fungi and aflatoxins occurrence. The analyzed feed types showing high *Aspergillus flavus* counts, but their isolates did not produce these metabolites. González, Pinto, and Felicio (2001) conducted a study from 1989 to 1999 with 728 food samples and found that 18% of them were contaminated with mycotoxins. In total, 338 of the total sample (n=728) were feed intended for livestock and domestic animals, and 17% of it had one or more mycotoxins, at varying levels, including some that did not comply with the Brazilian legislation, which is currently ruled by Collegiate Board Resolution (CBR) n. 138, from February 8, 2017 (Brasil, 2017).

Despite this subject's relevance of this subject given the relevance of fish farming to the country, information on fungi presence in fish remains scarce in Brazil (Atayde *et al.*, 2014). This economic activity is highly representative of Matanhão State's economic and social development, mainly because it is small rural producers' income source. This productive activity drives small municipalities economy by generating income and new job positions, mainly in the countryside - 'tambaqui' is the very basis of fish farming in this region. Therefore, the aforementioned aspects justify the aim of the current study, namely: assessing fungal contamination occurrence in *Colossoma macropomum* from fish farms on Maranhão Island.

## MATERIAL AND METHODS

### Ethical aspects

All methodological procedures complied with ethical principles set by the Brazilian College of Animal Experimentation and the Ethics Committee on Animal Experimentation (CEEAA) of State University of Maranhão (UEMA) (Protocol n. 09/2022).

### Collection site

Fish were collected from fish farms in Paço do Lumiar, Raposa, São José de Ribamar and São Luís municipalities, on Maranhão Island. They were labeled as P1, P2, P3 and P4. In total, 40 *C. macropomum* (tambaqui) specimens (male and female - at the fattening phase) were collected; 10 in each municipality and fish farm.

The specimens were captured with 2.5 mesh (25 mm) cast net and taken (still alive) to the Food and Water Microbiology Laboratory of State University of Maranhão (UEMA) in insulated boxes filled with water from the capture site, on the collection day. Subsequently, the collected specimens were euthanized by puncturing the upper part of their heads with a scalpel blade.

### Laboratory analysis

Fish were aseptically dissected in the laboratory with the aid of forceps and scissors to remove the longitudinal skeletal striated muscle (dorsal and ventral muscles) from all sampled fish. The removed fragments were placed in sterile plastic bags and analyzed right after.

Twenty-five ( $25 \pm 0.2$ ) grams of each specimen were weighed and added to 225 mL of 0.1% buffered peptone water, which corresponded to the first dilution ( $10^{-1}$ ), for microbiological analysis. This dilution was homogenized for approximately 2 minutes in stomacher at rotation speed of 250 revolutions per minute (rpm), on average. Subsequently, two more successive dilutions ( $10^{-2}$  and  $10^{-3}$ ) were prepared in test tubes added with 9 mL of 0.1% buffered peptone water.

### Filamentous fungi enumeration and identification

Pour plate plating, based on using 1 mL of serial dilutions ( $10^{-1}$ ,  $10^{-2}$  and  $10^{-3}$ ) on Potato Dextrose Agar (PDA) acidified with 10% tartaric acid solution was the technique adopted for filamentous fungi enumeration. The plates were incubated in oven at  $25 \pm 2$  °C for seven (07) days, in the dark, after the aforementioned procedure. Fungal colonies enumeration was carried out on plates presenting 10 to 100 CFU/g, according to Dalcero *et al.* (1997) and Dalcero *et al.* (1998). Results were expressed as Colony Forming Units (CFU) per gram.

Filamentous fungal colonies were selected, isolated and purified after enumeration through successive sub-culturing and plating on PDA medium. Subsequently, the developed fungi were analyzed and identified. Identification was based on both macroscopic morphology and colony features (obverse and reverse fungal growth on petri dishes), as recommended by Sidrim, Brilhante and Rocha (2004).

These features are (i) shape (circular or irregular), (ii) pigmentation (depending on color), (iii) texture (cottony, furfuraceous, downy, sandy or powdery, velvety, membranous, glabrous, creamy), (iv) rim (smooth, fringed, wavy, pleated, rowed, lobed, and filamentous), (v) size (small, medium and large - diameter in mm) and (v) luster (opaque, moderate and intense).

Microscopic morphology was assessed based on the identification keys recommended by Silveira (1995), Lacaz *et al.* (1998) and Kawashima, Soares and Massaguer (2002) to identify the isolated filamentous fungi at genus level.

## RESULTS AND DISCUSSIONS

The enumeration of filamentous fungi isolated from *C. macropomum* is summarized in Table 1, according to which, there was filamentous fungi in 67.5% (n= 27/40) of the assessed specimens - populations ranged from  $1.0 \times 10^1$  to  $4.52 \times 10^4$  CFU/gram.

**Table 1** – Filamentous fungi enumeration in *Colossoma macropomum* from fish farms on Maranhão Island.

| Samples | Filamentous Fungi Enumeration (UFC/g) |                   |                   |                    |
|---------|---------------------------------------|-------------------|-------------------|--------------------|
|         | P1                                    | P2                | P3                | P4                 |
| 1       | $3.0 \times 10^1$                     | $2.0 \times 10^1$ | $1.0 \times 10^1$ | <10                |
| 2       | <10                                   | $6.0 \times 10^1$ | $1.7 \times 10^2$ | <10                |
| 3       | $2.2 \times 10^2$                     | $1.0 \times 10^2$ | $6.0 \times 10^1$ | $1.81 \times 10^3$ |
| 4       | $3.6 \times 10^2$                     | <10               | $3.0 \times 10^2$ | $4.52 \times 10^4$ |
| 5       | <10                                   | $1.4 \times 10^3$ | $8.0 \times 10^1$ | $1.24 \times 10^3$ |
| 6       | <10                                   | $7.0 \times 10^1$ | $3.3 \times 10^2$ | $4.07 \times 10^4$ |
| 7       | <10                                   | <10               | $6.1 \times 10^1$ | $9.88 \times 10^3$ |
| 8       | $2.2 \times 10^2$                     | $1.0 \times 10^1$ | $2.4 \times 10^2$ | <10                |
| 9       | <10                                   | $3.7 \times 10^3$ | $5.1 \times 10^2$ | <10                |
| 10      | <10                                   | $1.0 \times 10^1$ | $1.4 \times 10^3$ | <10                |

Brazil does not have regulations aimed at enumerating fungi in fish, regardless of fish category (raw, seasoned or not, fresh, chilled or frozen). Similarly, there are no legal standards for fungi counting in animal feed. The standard adopted for animal feed certification (GMP, 2008) in other countries is based on counts that cannot exceed 4.00 CFU/g in log10 to ensure the product's hygienic quality. Therefore, based on this standard, 20 (74.07%) of the 27 samples recorded values above the limits set for animal feed, if one bears in mind the enumeration of each analyzed fish.

Fungal count results recorded for the 20 assessed specimens suggested high fungal population prone to spoil the fish and to lead to low microbiological quality raw material for compromising its organoleptic and nutritional features. According to Ribeiro *et al.* (2009), fungal contamination level can vary depending on the region's climatic conditions, collection period, food processing conditions and storage method.

Fungal diversity in *C. macropomum* consisted of 12 isolates based on macroscopic morphology and colonial features (Tabs. 2 and 3). These criteria showed that they presented typical morphologies (Sidrim; Brillhante; Rocha, 2004; Carvalho, 2013; Figueiredo *et al.*, 2020) used as preliminary fungal differentiation

method. Vecchia and Castilhos-Fortes (2007), Uwadiae, Ebonne (2011) and Cardoso Filho *et al.* (2013) also reported filamentous fungi identification based on macroscopic morphology and colony features.

Genera *Aspergillus* sp. (n= 07 isolates), *Penicillium* sp. (n= 03 isolates), *Paecilomyces* sp. (n= 01 isolate) and *Rhizopus* sp. (n= 01) were identified through microscopic morphology (Table 4) associated with macroscopic morphology. According to Araújo *et al.* (2010), *Aspergillus*, *Penicillium* and *Rhizopus* are classified as storage fungi. They take over the food substrate shortly before and during storage; therefore, they are found in large numbers in warehouses, mills, silos, hoppers, elevators, equipment and places where agricultural products are stored, handled and processed.

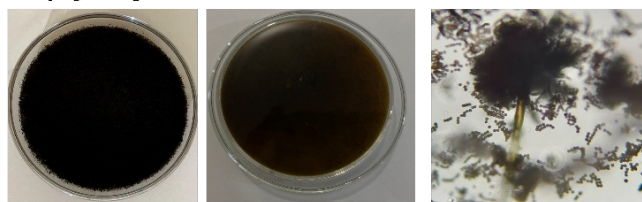
The fungal group most commonly contaminating the assessed fish belonged to genus *Aspergillus* sp. Its development, and macroscopic and microscopic features, can be observed in Figure 1 (A, B, and C). According to Carvalho (2013), in macroscopic terms, this fungi group colonies can macroscopically present white color at early maturation. The color can evolve to green, yellow, brown or black, depending on the species. Furthermore, it shows cottony texture. In microscopic terms, they have filamentous shape and septate hyphae.

**Figure 1** - Macroscopic (fungal obverse and reverse colonies in petri dishes added with potato dextrose agar, at seven-day growth) and microscopic view of filamentous fungi (40x microscopy photographs) belonging to genus *Aspergillus*: (A) *Aspergillus* sp.; (B) *Aspergillus niger*; (C) *Aspergillus terreus*.

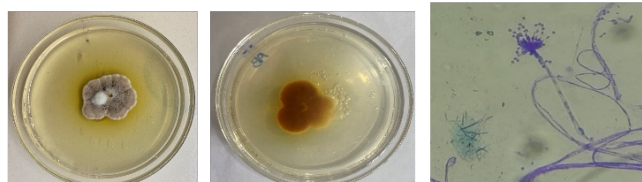
A - *Aspergillus* sp.



B - *Aspergillus niger*



C - *Aspergillus terreus*



Source: Authors' files.

The applied identification keys enabled determining two *Aspergillus* species: *A. niger* and *A. terreus*. According to Carvalho (2013), vesicle shape (spherical, hemispherical, elongated or elliptical) is one of the main features used to identify and differentiate one species from the other. Color is also necessary to help getting to a diagnosis.



**Table 2** – Filamentous fungi macroscopic morphology and features, obverse colonies, *Colossoma macropomum* isolated from fish farms on Maranhão Island.

| Isolated Fungi | Diameter (mm) | Obverse Colonies |              |         |                   |            |                        |        |          |              |
|----------------|---------------|------------------|--------------|---------|-------------------|------------|------------------------|--------|----------|--------------|
|                |               | Texture          | Pigmentation | Surface | Rim               | Topography | Color                  | Aspect | Pigment  | Growing time |
| 1              | 40.00         | Suede            | Unchanged    | Rough   | Uneven            | Pleated    | Green, yellow core     | Opaque | Absence  | Slow         |
| 2              | 73.00         | Powdery          | Unchanged    | Rough   | White             | Flat       | Black                  | Wet    | Absence  | Fast         |
| 3              | 75.00         | Powdery          | Unchanged    | Rough   | Regular           | Flat       | Black                  | Dry    | Absence  | Fast         |
| 4              | 60.00         | Velvety          | Unchanged    | Rough   | Regular           | Flat       | White, yellow core     | Wet    | Absence  | Slow         |
| 5              | 72.00         | Velvety          | Unchanged    | Rough   | Regular           | Flat       | Black                  | Opaque | Absence  | Slow         |
| 6              | 72.00         | Powdery          | Unchanged    | Rough   | White             | Flat       | Brown                  | Wet    | Absence  | Fast         |
| 7              | 21.00         | Velvety          | Unchanged    | Cracked | Regular and White | Flat       | Green with white core  | Wet    | Absence  | Slow         |
| 8              | 25.00         | Coriaceous       | Unchanged    | Cracked | Uneven and white  | Pleated    | Green                  | Opaque | Absence  | Slow         |
| 9              | 26.00         | Cotton           | Changed      | Cracked | Uneven            | Pleated    | White with yellow core | Wet    | Absence  | Fast         |
| 10             | 25.00         | Velvety          | Changed      | Cracked | Uneven and white  | Pleated    | Salmon with white core | Wet    | Presence | Slow         |
| 11             | 40.00         | Cotton           | Unchanged    | Cracked | Uneven            | Pleated    | White with black core  | Wet    | Absence  | Fast         |
| 12             | 21.00         | Cotton           | Changed      | Cracked | Uneven            | Umbilicate | White                  | Wet    | Absence  | Slow         |

**Table 3** – Filamentous fungi macroscopic morphology and features, reverse colonies, *Colossoma macropomum* isolated from fish farms on Maranhão Island.

| Isolated Fungi | Obverse Colonies |         |            |                                     |          |
|----------------|------------------|---------|------------|-------------------------------------|----------|
|                | Surface          | Rim     | Topography | Color                               | Pigment  |
| 1              | Cracked          | Uneven  | Pleated    | Brown                               | Absence  |
| 2              | Rough            | Uneven  | Pleated    | Yellow                              | Absence  |
| 3              | Smooth           | Regular | Flat       | Brown                               | Absence  |
| 4              | Smooth           | Regular | Pleated    | Yellow rim and red core             | Absence  |
| 5              | Smooth           | Regular | Flat       | Brown with white rim                | Absence  |
| 6              | Rough            | Regular | Flat       | Light brown                         | Absence  |
| 7              | Cracked          | Regular | Pleated    | Cream with white rim                | Absence  |
| 8              | Cracked          | Uneven  | Pleated    | Cream with white rim                | Absence  |
| 9              | Cracked          | Uneven  | Pleated    | Yellow with white rim               | Absence  |
| 10             | Smooth           | Uneven  | Flat       | Orange with white rim               | Presence |
| 11             | Cracked          | Uneven  | Pleated    | Cream with black core and white rim | Absence  |
| 12             | Cracked          | Uneven  | Pleated    | Yellow with white rim               | Absence  |

**Table 4** – Microscopic features of isolates from *Colossoma macropomum* collected from fish farms in São Luís Metropolitan Region – MA.

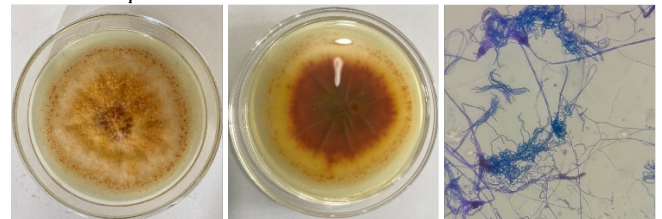
| Fungus | Hyphae                   | Conidiophore       | Vesicle                       | Phialides   | Conidia                       |
|--------|--------------------------|--------------------|-------------------------------|-------------|-------------------------------|
| 1      | Septate and Hyaline      | Smooth and long    | Fully covered                 | Compact     | Smooth, hyaline and spherical |
| 2      | Septate and Hyaline      | Smooth             | Fully covered                 | Compact     | Smooth, hyaline and spherical |
| 3      | Septate and Dematiaceous | Smooth and long    | Spherical                     | Irradiated  | Dematiaceous and spherical    |
| 4      | Septate and Hyaline      | Septate            | Absent                        | Vial-shaped | Smooth, hyaline and spherical |
| 5      | Septate and Hyaline      | Smooth and long    | Composed of up to 7 phialides | Elongated   | Oblong                        |
| 6      | Septate and Dematiaceous | Smooth and long    | Spherical                     | Irradiated  | Dematiaceous and spherical    |
| 7      | Septate and Hyaline      | Smooth and long    | Fully covered                 | Irradiated  | Smooth, hyaline and spherical |
| 8      | Septate and Hyaline      | Smooth and long    | Fully covered                 | Compact     | Smooth, hyaline and spherical |
| 9      | Septate and Hyaline      | Smooth and Hyaline | Absent                        | Vial-shaped | Smooth, hyaline and spherical |
| 10     | Septate and Hyaline      | Smooth             | Fully covered                 | Compact     | Smooth, hyaline and spherical |
| 11     | Septate and Hyaline      | Smooth             | Fully covered                 | Compact     | Smooth, hyaline and spherical |
| 12     | Lisa and Hyaline         | Smooth             | Fully covered                 | Vial-shaped | Smooth, hyaline and spherical |

According to Corbellini *et al.* (2003), *A. niger* is not associated with mycotoxins production, but it is related to fungal infections and miscarriage episodes in livestock, such as cattle. Blane, Loret and Goma (1995), Pier and Richard (1992), and Manabe (2001) reported that *A. terreus* produces secondary metabolites like patulin and citrinin, which are harmful to humans. Lass-Flörl *et al.* (2021) stated that *A. terreus* is a species complex of great interest nowadays because they are often resistant to amphotericin B. Recent epidemiological surveys showed higher *A. terreus* incidence, the expansion of its clinical spectrum (chronic infections) and, consequently, new groups of patients at risk of being affected.

Genus *Penicillium* sp. was the group taking the second position in the ranking of groups causing fungal contamination in the herein assessed fish. This group is very large and holds several species known for causing food spoilage and mycotoxins production (ochratoxin A, rubrosulin, viopurpurin, viomelin, citrinin, citreoviridin, islanditoxin, cyclopiazonic acid, and patulin) (Zain, 2011). Patulin, for example, is a mycotoxin capable of causing neurotoxic and immunological effects, as well as gastrointestinal alterations in animals (Kawashima; Soares; Massaguer, 2002; Rupollo *et al.*, 2004). It is found in stored products like cereals and in other environments, such as in the air (Mezzari *et al.*, 2002; Wang *et al.*, 2017).

The development of fungi belonging to genus *Penicillium* sp. in the assessed fish, and their macroscopic and microscopic features, can be observed in Figure 2 (A, B and C).

According to Figueiredo *et al.* (2020), based on its macromorphology, *Penicillium* colonies often show a wide variety of colors ranging from grayish-green, orange, yellow, blue, gray to white. Oftentimes, more than one color can be observed in a colony at the same time. When it comes to texture, colonies can be flocculent, velvety, fasciculate and simethicone. Conidiophores are classified as with solitary phialides, monoverticillate, biverticillate, divaricate, terverticillate

**Figure 2** - Macroscopic view of fungal obverse and reverse colonies in petri dishes added with dextrose agar, at seven-day growth) and microscopic views of filamentous fungi (40x microscopy photographs) belonging to genus *Penicillium* sp.**A - *Penicillium* sp.****B - *Penicillium* sp.**

Source: Authors' files.

and quaterverticillate.

Genera *Rhizopus* sp. and *Paecilomyces* sp. were also identified, in addition to *Aspergillus* sp. and *Penicillium* sp. According to Marcia and Lazzari (1998), both genera are commonly found in the soil, in vegetables, fruits and stored grains. They are common contaminants in places where stored products are processed.

Studies on fungal contamination in fish are noteworthy, as they allow acquiring information inherent to the quality of the raw material and about the need for implementing preventive actions at different production phases in order to control fungi and their secondary metabolites (mycotoxins), in addition to applying good agricultural practices to reduce food contamination and avoid losses in animal production, as well as risks to animal and human health.

## CONCLUSIONS

Based on the results, the analyzed *C. macropomum* specimens presented high fungal contamination by the identified genera, namely: *Aspergillus* sp., *Penicillium* sp., *Paecilomyces* sp. and *Rhizopus* sp. The first two of these genera are potentially mycotoxigenic.

Fish is a nutritionally rich seafood type presenting intrinsic features that favor their deterioration depending on the conditions provided by the sampled fish farms. These conditions can be favorable to fungi survival and multiplication, which requires more effective control and monitoring, including the storage of feed provided to the animals in fish farms. All these precautions aim at ruling out the likely occurrence of toxin-producing fungi that can compromise fish quality, have impact on production and cause health issues in consumers.

## REFERENCES

- ARAÚJO, A. G. S. de *et al.* Ocorrência de fungos de campo e de armazenamento em ingredientes e ração para tambaqui (*Colossoma macropomum*). **Pubvet**, v. 4, n. 35, p. 1-26, 2010.
- ARRUDA, A. D.; BERETTA, A. L. R. Z. Micotoxinas e seus efeitos à saúde humana: revisão de literatura. **Revista Brasileira de Análises Clínicas**, v. 51, n. 4, p. 286-289, 2019.
- ATAYDE, H. M. *et al.* Fungos toxigênicos e micotoxinas na alimentação de peixes: uma revisão. **Scientia Amazonia**, v. 3, n. 3, p. 59-71, 2014.
- BATISTA, L. R.; FREITAS, R. F. de. Avaliação da produção de aflatoxinas por espécies do fungo *Aspergillus* associados ao café. **Revista Brasileira de Armazenagem**, Viçosa, Especial, v. 1, p. 44-49, 2000.
- BLANE, P. J.; LORET, M. O.; GOMA, G. Production of citrinin by various species of *Monoascus*. **Biotechnology Letter**, v. 17, p. 291-294, 1995.
- BRASIL. Ministério da Saúde. Agência Nacional de Vigilância Sanitária. **Resolução da Diretoria Colegiada (RDC) nº 138, de 08 de fevereiro de 2017**. Altera a Resolução da Diretoria Colegiada - RDC nº 7, de 18 de fevereiro de 2011, que dispõe sobre limites máximos tolerados (LMT) para micotoxinas em alimentos, para alterar os LMT da micotoxina deoxinivalenol (DON) em trigo e produtos de trigo prontos para oferta ao consumidor e os prazos para sua aplicação. 2017. Disponível em: [https://bvsms.saude.gov.br/bvs/saudelegis/anvisa/2017/rdc0138\\_08\\_02\\_2017.pdf](https://bvsms.saude.gov.br/bvs/saudelegis/anvisa/2017/rdc0138_08_02_2017.pdf).
- CARDOSO FILHO, F. das C. *et al.* Monitoramento de fungos toxigênicos e aflatoxinas em rações utilizadas em piscicultura. **Ciência Animal Brasileira**, Goiânia, v. 14, n. 3, p. 305-311, 2013.
- CARVALHO, L. I. C. ***Aspergillus* e Aspergilose - desafios no combate da doença**. 2013. 56 g. Dissertação (Mestrado Integrado em Ciências Farmacêuticas) - Universidade Fernando Pessoa, Porto, 2013.
- CHIOTTA, M. L. *et al.* *Aspergillus* section Nigri species isolated from different wine-grape growing regions in Argentina. **International Journal of Food Microbiology**, v. 136, p. 137-141, 2009.
- CORBELLINI, L. G. *et al.* Aborto por *Aspergillus fumigatus* e *A. niger* em bovinos no sul do Brasil. **Pesquisa Veterinária Brasileira**, Rio de Janeiro, v. 23, n. 2, p. 82-86, 2003.
- DALCERO, A. *et al.* Mycoflora and incidence of aflatoxin B1, zearalenone and deoxynivalenol in poultry feeds in Argentina. **Mycopathologia**, v. 137, n. 3, p. 179-184, 1997.
- DALCERO, A. *et al.* Mycoflora and naturally occurring mycotoxins in poultry feeds in Argentina. **Mycopathologia**, v. 141, n. 1, p. 37-43, 1998.
- DANTIGNY, P.; GUILMART, A.; BENSOUSSAN, M. Basis of predictive mycology. **International Journal Food Microbiology**, v. 100, p. 187-196, 2005.
- FIGUEIREDO, C. N. *et al.* **Diversidade taxonômica e identificação de *Penicillium***. In: SOARES, A. C. F. *et al.* (Orgs.). **Tópicos em Microbiologia Agrícola**. Cruz das Almas, BA: EDUFRB, 2020. p. 247-266.
- FILHO, A. A.; CAMPOLINA, D.; DIAS, M. B. **Toxicologia clínica**. 2. ed. Folium: Belo Horizonte, 2013. 675 p.
- FRANCO, B. D. G. M.; LANDGRANF, M. **Microbiologia dos Alimentos**. 1. ed. Atheneu. São Paulo. 2008. 171 p.
- GMP (Good Manufacturing Practices). **Certification Scheme Animal Feed**. Sector 2008, Appendix 1: Product standards; Regulations on Product Standards in the Animal Feed Sector. GMP14, p. 1- 39, 2008.
- GONÇALEZ, E.; PINTO, M. M.; FELICIO J. D. Análise de micotoxinas no Instituto Biológico de 1989 a 1999. **O Biológico**, São Paulo, v. 63, n. 1/2, p. 15-19, 2001.
- GRIESSLER, K.; ENCARNAÇÃO, P. Fumonins - mycotoxins of increasing importance in fish! **Aquaculture Asia Magazine**, p. 24-26, 2009.
- HERMANN, G. *et al.* Fungos e fumonisinas no período pré-colheita do milho. **Ciência & Tecnologia de Alimentos**, Campinas, v. 26, n. 1, p. 7-10, 2006.
- KAWASHIMA, L. M.; SOARES, L. M. V.; MASSAGUER, P. R. de. The development of an analytical method for two mycotoxins, patulin and verruculogen, and survey of their presence in commercial tomato pulp. **Brazilian Journal of Microbiology**, São Paulo, v. 33, n. 3, p. 269-273, 2002.
- KNASMULLER, S. *et al.* "Structurally Related Mycotoxins Ochratoxin A, Ochratoxin B, and Citrinin Differ in Their Genotoxic Activities and in Their Mode of Action in Human-Derived Liver (Hep G2) Cells: Implications for Risk Assessment". **Nutrition and Cancer**, v. 50, n. 2, p. 190-197, 2004.
- LACAZ, C. S. *et al.* **Guia para identificação - fungos Actinomicetos - Algas de interesse médico**. 1. ed. São Paulo: SARVIER, 1998. 445 p.

- LASS-FLÖRL, C. *et al.* Complexo de Espécies *Aspergillus terreus*. **Clinical Microbiology Reviews**, v. 34, n. 4, p. e0031120, 2021.
- MANABE, M. Fermented foods and mycotoxins. **Mycotoxins**, v. 51, p. 25-28, 2001.
- MARCIA, B. A.; LAZZARI, F. A. Monitoramento de Fungos em milho em grão, grits e fubá. **Ciência & Tecnologia de Alimentos**, Campinas, v. 18, n. 4, p. 363-367, 1998.
- MEZZARI, A. *et al.* Fungos anemófilos na cidade de Porto Alegre, Rio Grande do Sul, Brasil. **Revista do Instituto de Medicina Tropical**, v. 44, n. 5, p. 269-272, 2002.
- PATERSON, R. R. M.; VENÂNCIO, A.; LIMA, N. Solutions to *Penicillium* taxonomy crucial to mycotoxin research and health. **Research in Microbiology**, v. 155, n. 7, p. 507-513, 2004.
- PEREIRA, M. M. G. *et al.* Aflatoxinas em alimentos destinados a bovinos e em amostras de leite da região de Lavras, Minas Gerais – Brasil. **Ciência e Agrotecnologia**, v. 29, n. 1, p. 106-112, 2005.
- PIER, A. C.; RICHARD, J. L. Mycoses and mycotoxicoses of animals caused by *Aspergilli*. In: BENNETT, J. W.; KLICH, M. A. **Aspergillus: biology and industrial applications**. Maryland: Butterworth-Heinemann, 1992. p. 233-267.
- RIBEIRO, J. M. M. *et al.* Radiação gama sobre a micobiota de ração avícola e *Aspergillus* spp. **Ciência Rural**, v. 39, n. 5, p. 1452-1458, 2009.
- RODRIGUEZ-AMAYA, D. B.; SABINO, M. Pesquisa em micotoxinas no Brasil: a última década em foco. **Brazilian Journal of Microbiology**, v. 33, n. 1, p. 1-11, 2002.
- ROSA, C. A. R. *et al.* Mycoflora of poultry feeds and ochratoxin-producing ability of isolated *Aspergillus* and *Penicillium* species. **Veterinary Microbiology**, v. 113, n. 1-2, p. 89-96, 2006.
- RUPOLLO, G. *et al.* Sistemas de armazenamentos hermético e convencional na conservabilidade de grãos de aveia. **Ciência Rural**, v. 34, n. 6, p. 1715-1722, 2004.
- SIDRIM, J. J. C.; BRILHANTE, R. S. N.; ROCHA, M. F. G. Aspectos Gerais de fungos filamentosos e dimórficos na apresentação filamentosa. In: SIDRIM, J. J. C.; ROCHA, M. F. G. **Micologia Médica à luz de autores contemporâneos**. Guanabara Koogan, Rio de Janeiro, p. 83-86, 2004.
- SILVEIRA, V. D. **Micologia**. 5. ed. Rio de Janeiro: Âmbito Cultural, 1995.
- SIMAS, M. M.; BOTURA M. B.; CORREA, B. Determination of fungal microbiota and mycotoxins in brewers grain used in dairy cattle feeding in the State of Bahia, Brazil. **Food Control**, v. 18, p. 404-408, 2007.
- TACON, A. G.; METIAN, M. Aquaculture feed and food safety. **Annals of the New York Academy of Sciences**, n. 1140, p. 50-59, 2008.
- UWADIAE; R. E.; EBONNE, J. Benthic fungal and molluscan assemblages of a tropical mangrove swamp: impact of sediment characteristics on biodiversity and ecosystem function. **Researcher**, v. 3, n. 9, p. 41-59, 2011.
- VECCHIA, A. D.; CASTILHOS-FORTES, R. Contaminação fúngica em granola comercial. **Ciência & Tecnologia de Alimentos**, v. 27, n. 2, p. 324-327, 2007.
- WANG, X. C. *et al.* Phylogeny and morphological analyses of species. **Scientific Reports**, v. 7, n. 1, p. 1-14, 2017.
- ZAIN, M. E. Impact of mycotoxins on humans and animals. **Journal of Saudi Chemical Society**, v. 15, p. 129-144, 2011.