Isolation of Fungus From The Cadaver Storage Pool

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Abstract

Fungi made from samples taken from the ambient air of the cadaver storage pool and anatomy laboratory containing formaldehyde, *Purpureocellium lilacinum, Penicillium verrucosum, Aspergillus flavus, Trichophyton verrucosum, Trichophyton tonsurans,* and *Fusarium spp.* isolated and identified. In these cases, those grown formaldehyde fungi, which are used for the fixation of cadavers and the preservation of their structures, show that they should be constantly controlled in the cadaver pools. In addition, it is emphasized that the detected fungal agents may pose a risk in terms of health due to their zoonotic character.

Keywords: Animal, cadaver, formaldehyde, storage, zoonotic fungi.

Kadavra Saklama Havuzundan İzole Mantar Türleri

Özet

Formaldehit içeren kadavra tanklarından ve anatomi laboratuvarının ortam havasından alınan örneklerden yapılan mantar analizleri sonucunda *Purpureocellium lilacinum*, *Penicillium verrucosum*, *Aspergillus flavus*, *Trichophyton verrucosum*, *Trichophyton tonsurans* ve *Fusarium spp*. izole ve identifiye edilmiştir. Bu olgu ile birlikte anatomi eğitiminde kullanılan kadavraların fikzasyonu ve yapılarının korunması için kullanılan formaldehitin, fungisidal etkinliğinin, havuzlarda sürekli kontrol edilmesi gerekliliğini göstermektedir. Ayrıca

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tespit edilen mantar etkenlerinin zoonotik karakterde olması yönüyle sağlık açısından risk teşkil edebileceği de vurgulanmaktadır.

Anahtar Sözcükler: Formaldehit, hayvan, kadavra, saklama, zoonotik mantar.

1. Introduction

Formaldehyde is a simple aldehyde with the molecular formula CH₂O, discovered by British chemist August Wilheld Von Hofmann in 1867. It is a colorless gas at room temperature, a chemical with flammable properties with an irritating odor (Elshaer and Mahmoud, 2017). Formaldehyde is a highly reactive component that is widely used in medicine, agriculture, and industrial processes as a disinfectant for killing bacteria and fungi, and is used in the fixation of cadavers in the field of anatomy, preservation of the cadaver's structure intact and fixation of tissues (Tišler and Zagorc-Končan, 1997; Ünsaldı and Çiftçi, 2010; Yu et al., 2015). Formaldehyde can prove to be toxic in any ways upon repeated exposure through inhalation or absorption through skin contact. The toxicity of formaldehyde is clinically seen with dermatitis in the contact area, irritant effects on the nasal, oral and pharyngeal mucosa, and mutagenic or carcinogenic effects, and causes bronchial asthma, ocular irritation, corneal clouding, leukaemia, congenital malformations and skin allergy (Kapoor and Sharma, 2020).

Animal cadavers are an essential teaching tool in the education of anatomists and veterinary students. Formaldehyde is widely used in anatomy applications in the fixation of cadavers, due to its antioxidant activity, protection of human and animal cadavers, and prevention of microbial growth. The most common problem with the fixation of animal cadavers with formaldehyde is the growth of microorganisms on the surface of the skin and organs (Al Aiyan et al., 2018). In some studies, it has been proven that some fungal agents can reproduce despite formaldehyde solution; *Aspergillus sydowii* HUA strain by Yu et al. (2015); in another study by Yu et al. (2014), *Aspergillus nomius* SGFA1 strain and *Penicillium chrysogenum* SGFA3 strain by Minemura et al. (Minemura et al., 2015). *Aspergillus oryzae* isolated and identified several formaldehyde-tolerant microorganisms, including *Trichoderma spp.* by Sawada et al. (Swada et al., 2006), and *Paecilomyces spp.* by Kondo et al. (2008).

The aim of this study was to investigate zoonotic fungi in cadaver storage pools kept for use in veterinary anatomy courses.

2. Materials and Methods

2.1. Sample

In our study, swab samples were taken from cadaver storage pool with sterile swabs on the growth of formaldehyde solution (14 %) in the cadaver storage pool at the anatomy laboratory.

2.2. Isolation and Identification

In addition, to examine the environment of the anatomy laboratory, Sabouraud Dextrose Agar (SDA) media were kept open for approximately 10 minutes in suitable places (Afshari et al., 2013; Rostami et al., 2016; Sahab et al., 2019). The suspicious specimens brought to the microbiology laboratory were inoculated into SDA medium and incubated in an oven at 25°C (Walsh et al., 2018). The incubated samples were examined on the 7th and 14th days. Colonies, where growth occurred in the media were determined macroscopically. Macroscopic morphologies of fungal colonies growing on SDA were examined both from the surface and reverse (Figure 1).



Figure 1. Surface macroscopic morphology of fungal agents grown on SDA medium 1. Aspergillus flavus 2. Fusariumspp. 3. Penicillium verrucosum 4. Purpureocellium lilacinum 5. Trichophyton verrucosum 6. Trichophyton tonsurans. Reverse macroscopic morphology of fungal agents grown on SDA medium 7. Aspergillus flavus 8. Penicillium verrucosum 9. Purpureocellium lilacinum 10. Trichophyton tonsurans 11. Trichopyton verrucosum

Samples from fungal colonies were transferred onto a clean slide and examined microscopically using lactophenol cotton blue (Figure 2).

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Figure 2. Microscopic images of fungal agents with X400 and X1000 magnification. 1. Aspergillus flavus 2. Fusarium spp. 3. Purpureocellium lilacinum 4. Microscopic images of Purpureocellium lilacinum with X1000 magnification. Microscopic images of fungal agents with X400 magnification. 5. Trichophyton vertucosum 6. Penicillium verticosum 7. Trichophyton tonsurans.

Isolated fungal species were identified (Kidd et al., 2016; Samanta, 2015; Walsh et al., 2018).

3. Results and Discussion

Purpureocellium lilacinum, Penicillium verrucosum, Fusarium spp. and *Trichophyton verrucosum* were identified from the cadaver pool. *Trichophyton tonsurans* were identified from the ambient air. It was determined that the agent isolated from both the cadaver pool and the ambient air was *Aspergillus flavus*.

Purpureocellium lilacinum, (formerly *Paecilomyces lilacinus*) is isolated from soil, rotting vegetation, insect, nematodes, and laboratory air (as a contaminant) and it causes keratitis, endophthalmitis, subcutaneous infection, osteomyelitis, pneumonia in immunocompromised patients and invasive sinusitis in the immune-sufficient and diabetic state in humans and other vertebrates (Kidd et al., 2016; Luangsa-Ard et al., 2008; Walsh et al., 2018). The *Fusarium* species are opportunistic pathogens that spread in aquatic and other organisms worldwide, causing hyalo hifomycosis, mycotic keratitis, onychomycosis, and infectious cause in aquatic animals such as sea horses, dolphins and aquatic animals (Cafarchia et al., 2020; Kidd et al., 2016; Sáenz, 2020). *Aspergillus flavus* is a species that is distributed worldwide and is found in normal soil and many materials. In humans and animals, it causes otitis, keratitis, acute and chronic invasive sinusitis, and those with pulmonary and systemic infection in immuno-compromised patients (Kidd, 2016).

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Ochratoxin A, which is nephrotoxic, carcinogenic, genotoxic, and immuno toxic for humans and animals, is produced by *Penicillium verrucosum*. Pathogenic *Penicillium* species are rare in humans, but they can cause otomycosis, endocarditis, and mycotic keratitis. *Trichophyton tonsurans* is an anthropophilic fungus species that spread worldwide and causes *tinea capitis* and *tinea gladiatorial* in humans (Kidd, 2016; Samanta, 2015). *Trichophyton verrucosum*, which causes ring worm in cattle, is a zoophilic fungus. Infection in humans occurs as a result of direct contact with cattle or infected fomites and is usually observed on the scalp, beard, and exposed areas of the body (Kidd, 2016). Bovine ring worm causes economic losses due to its unfavourable impact on animal development, meat, and milk production as well as leather quality. Due to the high zoonotic character of the disease, farmers and their families, veterinarians, artificial insemination operators and technicians, as well as everyone working in this region are at risk of infection (Agnetti et al., 2014).

4. Conclusions

Although different methods are preferred for the fixation and preservation of cadavers that are used extensively in Veterinary Anatomy application courses, the fixation method with formaldehyde is widely used. However, because formaldehyde is a volatile gas at room temperature, it loses its effectiveness as sublimation, and therefore its effectiveness must be constantly monitored and formaldehyde should be added to cadaver pools in a controlled manner. In addition, formaldehyde has many harmful effects, especially carcinogenic, mutagenic and allergic. The types of fungi identified in this study pose a risk to students and the academicals and technicals and technical staff and all workers in the department and the faculty. For these reasons, it should be preferred to follow the effectiveness of formaldehyde used for the fixation and preservation of cadavers in anatomy application courses, to protect cadavers with methods that are not harmful to health and/ or may cause less harm. The disadvantages of these methods include the harm caused by chemicals and waste, to human health, the environment and nature, as well as the need for high economic costs to ensure the continuity of chemicals needed to preserve cadavers decayed. Nowadays, the increasing use of developing technology in education and training is becoming mandatory to eliminate or reduce these damages.

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Ethic Statement: This study does not present any ethical concerns.

Conflict of Interest: The authors declared that there is no conflict of interest.

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