Acta Aquatica Turcica

E-ISSN: 2651-5474

Home Page: https://dergipark.org.tr/actaquatr 18(3): 359-368, 2022 DOI: 10.22392/actaquatr.1074940

Research Article **Araştırma Makalesi**

Little Known Aspects of Aquatic Insects: Myiasis

Sucul Böceklerin Az Bilinen Yönleri: Miyazis

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Received: 17.02.2022 **Accepted:** 21.03.2022 **Published:** 01.09.2022

How to Cite: Gökçe, D. (2022). Little known aspects of aquatic insects: Myiasis. *Acta Aquatica Turcica*, *18*(3), 359-368. https://doi.org/10.22392/actaquatr.1074940

Abstract: Among invertebrates, Diptera, an aquatic insect, has the largest group of species. Aquatic Diptera larvae live in a highly distinctive environment in contact with vertebrates, humans, contaminated water, and depositing eggs in the host organism due to their life cycle. This study aims to describe various aspects of *Clogmia albipunctata* one of myiasis insects causing a disease that affects both living and dead vertebrates as well as humans and whose symptoms are often overlooked. Furthermore, the study is remarkable since it is the first report of *C. albipunctata* (Psychodidae) in an indoor drainage system, except for humans, vertebrates, and natural ecosystems. SEM images gave a detailed description of the larvae and confirmed the species identification. When their life cycles were investigated, it was determined that in addition to myiasis, *C. albipunctata* larvae (drain fly or moth fly) played a vital role in the movement of bacteria from drains to indoor places, such as toilets, bathrooms, showers, and kitchens. Multi-drug resistant bacteria populate *C. albipunctata*, which possesses synanthropic behavior, and may play a major role in its transmission. This study focused on accidental myiasis.

Keywords

- Aquatic Diptera
- *Clogmia albipunctata*
- Myiasis
- Psychodidae
- Turkey

Özet: Omurgasızlar arasında Diptera, en fazla sucul türe sahip olan böcek grubudur. Sucul Diptera'nın yaşam döngüsünde omurgalılar ve insanlarla karşılaşması, konakçı organizmaya yumurtlaması veya yumurta bırakılan kontamine su ile olan temas, sucul Diptera larvalarının çok farklı bir çevrede yaşamasına sebep olur. Bu çalışmanın amacı, insanlar kadar canlı ve ölü omurgalıları etkileyen ve semptomları sıklıkla gözden kaçan miyaz böceklerinden biri olan *Clogmia albipunctata*' yı çeşitli yönleriyle tanımlamaktır. Ayrıca çalışma, *C. albipunctata*' nın (Psychodidae) insan, omurgalılar ve doğal ekosistemler dışında bir kapalı gider sistemindeki ilk kayıt olması nedeniyle dikkat çekicidir. SEM görüntüleri, larvanın detaylı tanımlamasına izin vermiş ve tür tanımlamasını doğrulamıştır. Yaşam döngüleri incelendiğinde miyazise ek olarak *C. albipunctata* larvalarının (gider sineği ya da güve sineği), bakterilerin giderlerden tuvalet, banyo, duş, mutfak gibi iç mekanlara taşınmasında kritik bir rol oynadığı görülmüştür. Ayrıca, çoklu ilaca dirençli bakteriler, sinantropik davranışa sahip olan ve bulaşıda önemli bir rol oynayabilen *C. albipunctata*' da yerleşir. Bu çalışma, sucul böceklerin tesadüfi miyazisi üzerine odaklanmıştır.

Anahtar kelimeler

- Sucul Diptera
- *Clogmia albipunctata*
- Miyazis
- Psychodidae
- Türkiye

1. INTRODUCTION

The population dynamics of freshwater benthic macroinvertebrates alter over time, depending on water quality and ecosystem productivity. Benthic macroinvertebrate community composition and ecological tolerance values of those invertebrate species based on environmental resistance provide important information for aquatic biomonitoring. Therefore, invertebrates have a crucial position in

aquatic ecology studies. Diptera, one of the aquatic insects, has the largest group of species among invertebrates. In its life cycle, aquatic Diptera encounters vertebrates and humans and contacts with contaminated water or laying eggs in the host organism, which leads Diptera larvae to live in a very different habitat and to be identified under different bio-ecological conditions: myiasis.

The term myiasis was first used by Hope in 1840 (El-Dib et al., 2020). Myiasis is defined as the infestation of human and vertebrate animals with insect larvae that feed on the host's dying (necrotic) or alive tissue, liquid body substances, or swallowed food for at least a period of time (El-Badry, 2014; El-Dib et al., 2020; Gökçe, 2020). As insect larvae, Diptera, Lepidoptera, Hymenoptera, and Coleoptera larvae cause myiasis (Cordeiro and Wagner, 2018). Myiasis is classified in two ways: anatomically, according to the location of the infestation on the host, and parasitically, according to the parasite's level of dependence on the host (Boumans et al., 2009; Hovius et al., 2011; Amro et al., 2018).

Myiasis is a condition in which invertebrate (especially Diptera) larvae infest the tissue and organ cavities of people and vertebrates, and lesions occur since the larvae feed with living or dead tissues, body fluids, or undigested food (Gökçe, 2020). Especially Calliphoridae, Sarcophagidae, and Destridae are groups that cause mostly myiasis in Diptera. Also, Fanniidae, Muscidae, Phoridae, Syrphidae, Psychodidae (Diptera) are crucial families that are responsible for myiasis worldwide (Ježek and van Harten, 2009; Gökçe, 2020).

Obligatory, facultative, and accidental myiasis are the three types of myiasis (Zittra et al., 2020; Mokhtar et al., 2016). There are two causes of accidental myiasis. The first is ingesting food contaminated with larvae. The second is when flies lay their eggs in either the host's anus or their urogenital area, thus causing the larvae to enter the rectum or urogenital tract. Nevertheless, the majority of the digested larvae are unable to complete their life cycle in the digestive or urogenital systems of their hosts. Cutaneous, subcutaneous, or cavitary groups are seen in myiasis according to the habitation of the attached larvae (Mohammed and Smith, 1976; Hjaija et al., 2018; Sarkar et al., 2018; El-Dib et al., 2020). Human myiasis is most commonly found in open wounds that have not been cared for properly. Furthermore, it can also affect body orifices including the oral cavity, eyes, ears, anus, and urogenital tract. Urogenital myiasis is a condition in which fly larvae infest the urinary canal and genital organs like the vaginal or penile orifices (Rasti et al., 2016; Hjaija et al., 2018; Pijáček M, Kudělková, 2018).

Mature flies are seen between the late prevernal and serotinal seasons and they lay ova. On the other hand, some myiasis agents are larvae inhabiting in aquatic habitats. The prevalence and frequency of myiasis are determined by fly and susceptible animal populations, as well as climate and environmental conditions (Kvifte and Wagner, 2017).

This study aims to describe different aspects of myiasis disease which affects both live and deceased vertebrates and human beings, but whose symptoms are frequently disregarded. Furthermore, the study is important because it is the first record about *Clogmia albipunctata* (drain fly or moth fly) as the habitat in Turkey, except for humans, vertebrates, and natural aquatic ecosystems.

2. MATERIAL and METHODS

2.1. Sampling and identification

In this study, larvae samples were collected around the sink and the drain filter in the building on the university campus in Malatya. Organic matter residue on the body of larvae prevents microscopic examination and clear SEM images. For the preparation of the specimens, a 10% KOH solution was utilized. The specimens were kept in 10% KOH solution at room temperature for 4 hours for cleaning from organic matter residue on the body of the larvae. Larvae were not left in the solution for a longer period of time to avoid degeneration of the soft portions of the body and the integrity of the body. After that, specimens were washed with distilled water and were preserved in 80% ethyl alcohol and glycerol solution. The identification process was performed according to Kvifte and Wagner (2017), Cordeiro and

Wagner (2018) by using a stereomicroscope (Leica MZ7.5). The samples were photographed, and their morphological measurement was performed using Leica camera DFC295 (Leica Application Suite, LAS version 4.5LAS). Scanning electron microscope images (SEM; LEO EVO-40xVP) were taken by Laboratory (İnönü University Scientific and Technology Research Centre).

3. RESULTS

Larvae were collected around the indoor drain filter of a building in the university campus. A total of 42 specimens were identified as aquatic larvae, *Clogmia albipunctata* Williston, 1893 (Diptera: Psychodidae) in the area with wet and partially organic materials.

All of the specimens were at the 4th instar stage. The body lengths ranged from 5.120 to 6.10 mm (Figure 1.). The body has 26 pseudo segments (annuli), is covered with well-sclerotized light brown color tergal plates; and one of the remarkable characteristics is the bristly body.

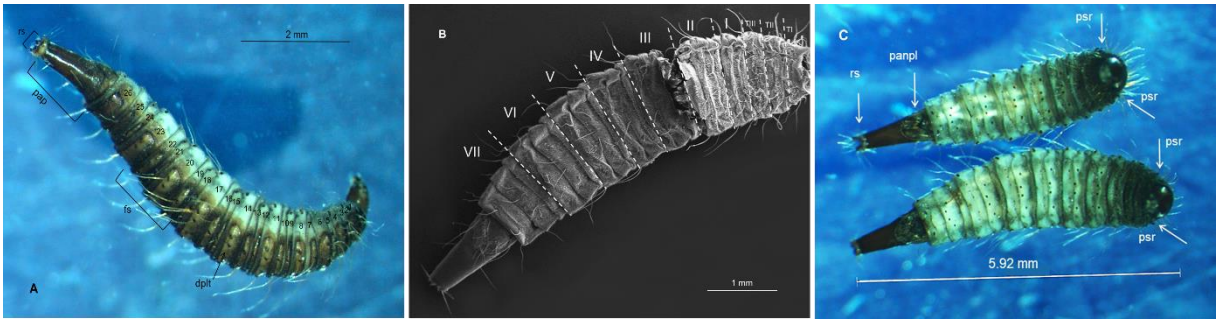

Figure 1. (A) *C. albipunctata* has totally 7 segments that are secondarily divided into 26 pseudosegments. (B) bristly 7 abdominal and 3 thoracic segments have filiform setae dorsolaterally view (65 X). (C) spiraculum and respiratory siphon are clearly recognizable in the whole body ventral view (*dplt:* dorsal plate; *fs:* filiform seta; *panpl:* preanal plate; *pap:* post abdominal process; *psr:* prothoracic spiracle; *rs:* respiratory siphon).

The head capsule is sub-oval and sclerotized. The hypostome has three teeth (Figures 2. and 3). The thorax is covered with tooth-like scales spination (Figure 3.).

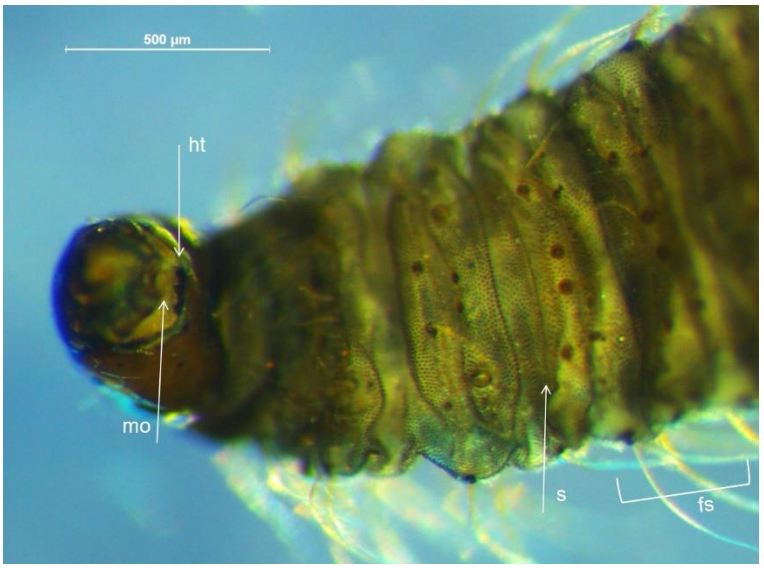

Figure 2. *C. albipunctata* larva has well development and sclerotized head capsule. Hypostomal three sharp teeth are prominent (*fs:* filiform seta; *ht:* hypostomal teeth; *mo:* mouth opening; *s:* spines).

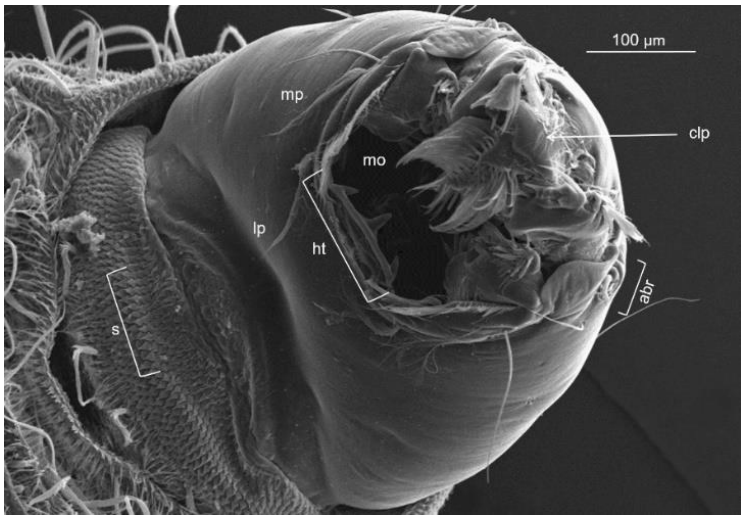

Figure 3. Head with the left antenna from a ventral view. Hypostomal teeth are clearly visible, median tooth slightly longer than corner teeth (550 X) (*abr:* antennal basal ring; *clp:* clypeus; *ht:* hypostomal teeth; *lp:* labial palpus; *mo:* mouth opening; *mp:* maxillary palpus; *s:* spines).

Prothoracic spiracles are present (Figure 4.). At the terminal end of the body, the respiratory siphon plate is prominent.

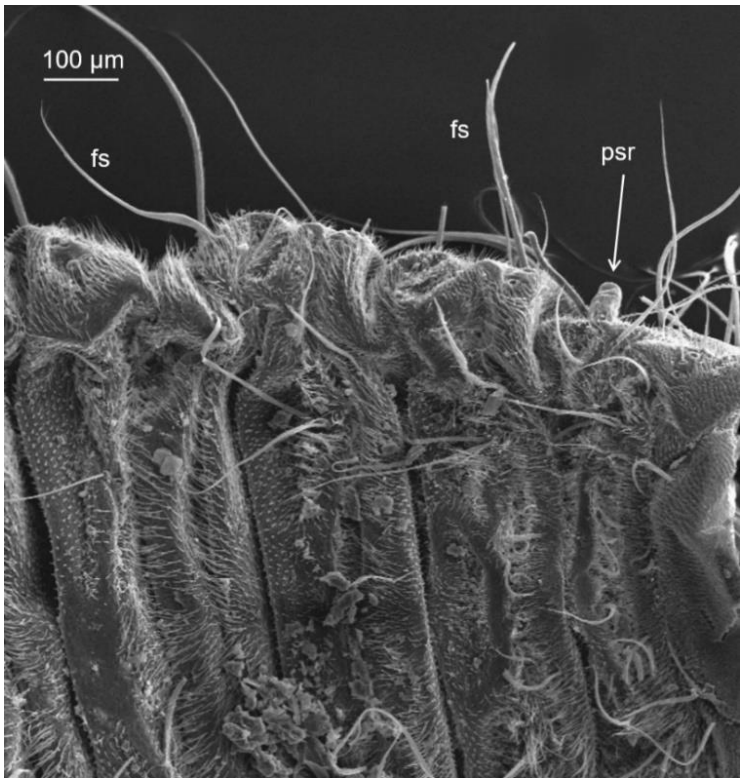

Figure 4. Prothorax has two finger-like spiracles in lateral positions, ventral view (250 X) **(***fs:* filiform seta; *psr:* prothoracic spiracle).

The preanal plate of *C. albipunctata* has a distinctive form (Figure 5.). SEM images were used to provide a detailed larval description and validation of the identification. After the filter which had found the larvae around it, was cleaned and covered, the larvae were not observed again. Also, no adult specimens were recorded.

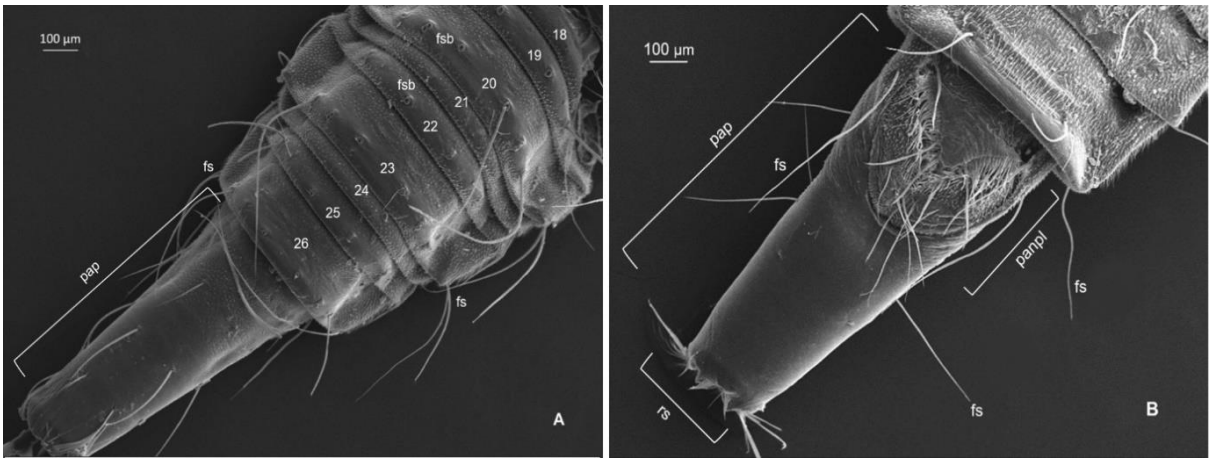

Figure 5. (A) *C. albipunctata* larval post abdominal part from dorsal view (210 X); (B) respiratory siphon plate from ventral view (230 X) (*fs:* filiform seta; *fsb:* filiform seta base; *panpl:* preanal plate; *pap:* post abdominal process; *rs:* respiratory siphon).

As in family characteristics, *C. albipunctata* has a four-stage life cycle as a holometabolous fly: egg, four larval instars, pupa, and adult. *C. albipunctata* is a synanthropic and cosmopolite aquatic Dipteran species (Wagner, 2011). The female ones lay their eggs on the surface of the water. Since they accumulate in higher numbers in indoor and outdoor wastewater pipe systems, they are commonly seen in wet bathrooms, hospitals, and drains (Ledwoch et al., 2018). They feed on the biofilm layer (protozoa, bacteria, algae) in pipes and drains (Ledwoch et al., 2018; Faulde and Spiesberger, 2013). Larval development depends on the amount of food and temperature. Adults and larvae are harmless. However, due to synanthropic life, larvae cause myiasis in vertebrates and humans.

The subfamily Psychodinae has previously been recorded in the Nearctic, Neotropic, Oriental, Afrotropic, and Australian zoogeographic regions (Wagner and Andersen, 2007). As a cosmopolitan species, *C. albipunctata* is common in most of the zoogeographic areas. Table 1 shows myiasis and zoogeographic location records of the species as larvae and adults. It can be regarded as an invasive species according to Oboňa and Ježek (2012).

Table 1. As a cosmopolitan species, *C. albipunctata* is widespread in most of the zoogeographic areas. Myiasis and zoogeographic field records of the larvae and adults are presented.

Life stage	Location	Country	References
Larvae	Intestinal myiasis	Japan	Tokunaga, 1953
Adult	Geographical location	Italy	Sarà and Salamanna, 1968
Larvae	Nasopharyngeal myiasis	Nigeria	Mohammed and Smith, 1976
Larvae	Urinary and intestinal myiasis	Malaya	Smith and Thomas, 1979
Adult	Geographical location	Senegambia	Wagner, 1983
Adult	Geographical location	Colombia	Wagner and Joost, 1994
Adult	Geographical location	Nicaragua	Maes and Killick-Kendrick, 1994
Adult	Geographical location	Germany, Central Europa	Werner, 1997
Adult	Geographical location	Tanzania,	Wagner and Andersen, 2007
Larvae	Intestinal myiasis	Taiwan	Tu et al., 2007
Adult	Geographical location	Mexico	Ibáñez-Bernal, 2008
Adult	Drain, hospital operation room	Belgium	Boumans et al., 2009
Adult	Geographical location	Arabian Peninsula and UAE	Ježek and Harten, 2009
Adult	Geographical location	Czech Republic	Wagner, 2011
Larvae	Urogenital myiasis	Germany	Hovius et al., 2011
Adult	Geographical location	Slovakia	Oboňa and Ježek, 2012
Larvae	Urinary myiasis	Egypt	El-Badry et al., 2014
Adult	Geographical location	Honduran	Bravo et al., 2014
Adult	Geographical location	Spain	Kvifte et al., 2016
Adult	Geographical location	Uruguay	Martinez et al., 2016
Larvae	Intestinal myiasis	Malaysia	Mokhtar et al., 2016
Adult	Geographical location	Thailand	Kvifte and Andersen, 2016
Larvae	Urinary myiasis	Iran	Rasti et al., 2016
Adult	Geographical location	Netherlands	Ciliberti et al., 2017
Larvae	Urinary myiasis	Israel	Sarkar et al., 2018
Larvae	Urinary myiasis	Palestine	Hjaija et al., 2018
Larvae	Drain	Venezuela	Cazorla-Perfetti, 2019
Larvae	Urogenital myiasis	Libya	Amro et al., 2019
Adult	Geographical location	Finland	Salmela et al., 2019
Adult	Geographical location	Austria	Zittra et al., 2020
Larvae	Intestinal myiasis	Egypt	El-Dib et al., 2020
Larvae	Urinary myiasis	Czech Republic	Pijáček and Kudělková, 2020
Larvae	Urogenital and gastrointestinal myiasis	Turkey	Gökçe, 2020
Adults	Geographical location	Ukraine	Oboňa et al., 2021
Larvae	Human residual root myiasis.	China	Liu et al., 2021
Larvae	Drain, Department flat, Academic facility	Turkey	Present study

4. DISCUSSION

Considering their life cycles, *C. albipunctata* larvae as well as myiasis play an important role in transporting bacteria since they move into indoor spaces through drains such as toilets, bathrooms, showers, and kitchen drains. It was noted by Faulde and Spiesberger (2012) that 45 bacterial species were isolated from the larvae of *C. albipunctata* collected in a hospital. Since *C. albipunctata*, which has a synanthropic behavior, is colonized by multi-drug resistant bacteria, it may play a crucial role in the transmission and contamination of multidrug-resistant bacteria that cause serious nosocomial infections. This relationship between bacteria and larvae often occurs in the environments such as hospitals and schools. The eggs and larvae pose a dangerous threat because they live in the biofilm contaminated with the patient's bacterial flora. The biofilm develops and spreads rapidly and can span distances of many kilometers. During the third and fourth larval stages, the larvae living in the biofilm may begin to move and thus can come out of damp areas such as showers, bathtubs, toilets, and kitchens. At this point, it can carry drug-resistant bacteria from the microbial flora of the biofilm to the environment (Rupprecht et al., 2020).

The emergence of *C. albipunctata*, on the other hand, primarily indicates inadequate water and pest management and sanitation in hospitals and other facilities (Faulde and Spiesberger, 2012). *Bacillus thuringiensis* is often regarded as the most effective larvicidal agent. It is frequently utilized as a microbiological agent against the world's most common insect pests. *B. thuringiensis* is known for producing a wide range of insecticidal proteins. According to Houston et al. (1989), the application of *Bacillus thuringiensis* serotype *israelensis* can reduce the incidence of drain flies by 79%.

Myiasis cases in Turkey were found to be caused by Diptera. Species belonging to the family Calliphoridae (Şenel et al., 2016), Sarcophagidae (Yücesan et al., 2021), Oestridae (Erenler et al., 2019), Psychodidae (Şahin et al., 2018; Gökçe, 2020; Şen and Polat, 2021), and Simuliidae (Akarsu et al., 2003) were recorded as the causative agent of myiasis in Turkey. These species are mostly aquatic Diptera larvae (Psychodidae and Simuliidae). Myiasis has become more common in rural regions due to sociocultural patterns and poor sanitation. This study focused on a different aspect of aquatic insects and described the 4th instar *C. albipunctata*, the first record in the drain in Turkey, in detail, and presented it to attract attention to myiasis which is usually overlooked.

5. CONCLUSION

This study revealed that all of the myiasis cases in Turkey is caused by synanthropic Dipterous larvae. Ecological factors such as temperature, nutrients, and moist conditions influence larval growth. Climatic change is a serious point as much as personal hygiene, and the spread and prevalence of accidental myiasis affect environmental health.

Today, two problems (low water quality and water scarcity), affect water consumption all over the world. At this point, an increase in the number of myiasis agents can be seen in aquatic insects due to low sanitation. In addition, there is an increase in the development of Dipterous larvae in the biofilm layer in drains and wastewater channels in indoor and outdoor environments. Along with its effect, myiasis creates serious health concerns by transmitting resistant pathogenic bacteria. The more eggs that get laid on the biofilm layer due to an increase in temperature exacerbate the insect invasion. For environmental health, disinfection processes that will leave minimum residue and ensure that other natural populations are minimally affected should be carried out. It is advised to provide regular drain cleaning to prevent hospital infections.

ACKNOWLEDGEMENT

The author would like to thank İnönü University Scientific and Technology Research Centre, SEM Laboratory for taking the Scanning Electron Microscope (SEM) images.

FUNDING

No financial support was received for the present study.

CONFLICT OF INTEREST

The author declares that has no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

AUTHOR CONTRIBUTIONS

DG is the corresponding author for the present study in all processes of manuscript preparation and final draft.

ETHICAL APPROVAL STATEMENTS

Local Ethics Committee Approval was not obtained because experimental animals were not used in this study.

DATA AVAILABILITY STATEMENT

Data supporting the findings of the present study are available from the corresponding author upon reasonable request.

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