pISSN: 0259-9996 – eISSN: 2783-3968

Journal of Entomological Society of Iran

2025, 45 (1), 97–106

Research Article

Species diversity of saprophagous flies (Diptera) in hospital grounds of Tehran, Iran

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Abstract. Synanthropic flies are essential in transmitting various pathogens, producing myiasis, and their application in forensic entomology. The main objective of this study was to identify the medically important flies in the hospital grounds of Tehran. The sampling process for this cross-sectional study was carried out every two weeks on the grounds of four hospitals in Tehran, using bottle traps and insect nets from March 2020 to February 2021. The population data has been transformed by calculating square root due to huge variations among community data, and data was analyzed with Kruskal-Wallis test to estimate the temporal and spatial variation. A total of 18,613 adult flies were collected and 18 species were identified. These flies belonged to four families: Calliphoridae, Muscidae, Sarcophagidae, and Fanniidae. The most common species in each family were *Calliphora vicina* Robineau-Desvoidy, *Muscina stabulans* Fallen, *Sarcophaga Africa* Wiedemann, and *Fannia canicularis* L., respectively. Temporal variations showed that *C. vicina* had different seasonal activity, with peaks in colder months, while *Lucilia sericata* Meigen, *Chrysomya albiceps* (Wiedemann), and dominant species of Sarcophagidae peaked in mid-summer. Spatial distribution analysis revealed overall unity in collecting all three species of Calliphoridae in all hospital environments. The same status was observed for Muscidae and Fanniidae. The dominant species in all species of the family Sarcophagidae were active during warmer months. *Sarcophaga nigriventris* Meigen, 1826 is reported for the first time for Iranian fauna.

Keywords: Nosocomial Myiasis, Forensic Entomology, Adult fly Trap

Article history
Received: 29 August 2024
Accepted: 11 December 2024
Published: 10 January 2025

Subject Editor: Ali Reza Sazmand

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DOI: https://doi.org/10.61186/jesi.45.1.8

Introduction

Flies of the order Diptera can affect human health. Most flies aren't harmful, but about 11000 species can produce diseases with various methods (Flieschmann et al 2004). Synanthropic flies are considered a global health problem and their impact on human health is a subject of discussion (Blunt et al 2011). Feeding strategy, inoculated environment, and their other behaviors changed them as the best candidates for mechanical transmissions of protozoan (Thaddeus et al 2005), viral, bacterial, and fungal infections to humans (Gupta et al 2012). Their larvae can also invade human and animal tissues, mucosa, and digested food and make various kinds of myiasis disease (Zumpt 1965).

The ability of mechanical transmissions of pathogenic agents by members of various true fly families such as Muscidae, Calliphoridae, Sarcophagidae, and Fanniidae has been proved (Greenberg 1973; Service 2014). Scientific studies on the potential mechanical transmission of pathogenic agents in some hospitals of Iran focused on house fly, *Musca domestica* (Diptera: Muscidae) (Kassiri et al 2012; Davari et al 2010) but various species of other families can be seen in hospital environments.

Infecting myiasis in patients after hospitalization is called nosocomial myiasis (Mielke 1997). Special conditions in hospitalized patients with weak bodies, the presence of open wounds or necrotic tissues, as well as mental disorders, enhance the risk of nosocomial myiasis in hospital environments (Dutto and Bertero 2010). Hospital environments could be attractive for necrophagous flies due to the presence of infectious disposal materials. The presence of medically important flies around hospital environments has been studied in Bandar Abbas City, Hormozgan province in south Iran (Sanei-Dehkordi et al 2020). Various kinds of human myiasis have been reported from Iran but reports on nosocomial myiasis are not frequent (Akbarzadeh et al 2012; Alizadeh et al

2014). Reporting a deadly myiasis case in Tehran in 2011 is clear evidence of the health-threatening effects of this kind of hospital-acquired myiasis (Mowlavi et al 2011). The main objective of this study was to identify the medically important flies in hospital environments of Tehran, the capital of Iran. Despite their importance in producing nosocomial myiasis, the data on the distribution of the flies can be used to complete the database of fly fauna of Iran which is applicable in forensic entomology investigations.

Materials and methods

Tehran is the capital of Iran, with 8.5 million people. At the sampling time, the mean temperature was between 5 - 35.5°C, and the mean humidity was between 30 – 62%. The city is located in a semi-arid area between Alborz Mountain in the north, and the central desert in the south. Its elevation ranged from 1200 to 1980 m from the lowest to highest point. Its weather is mild in spring and autumn, hot and dry in the summer, and cold in the winter.

Four general hospitals in the city were sampled (Fig 1). The selected hospitals are more popular and have a proportionally vast area. They are Taleqani Hospital (35° 47′ N, 51° 24′ E), with a 42000 m2 area, Farabi Hospital (35°67′ N, 51°39′ E), with a 17500 m2 area, Emam Hosein Hospital (35°70′ N, 51°45′ E), with a 93000 m2 area, and Shariat Razawi Hospital (35°67′ N, 51°32′ E), with a 13500 m2 area. The main method for collecting flies was bottle trap collection (Shiravi et al 2011). One piece of 25 gr of cow meat was inserted into each bottle trap to act as bait (Fig 2). Eight traps were installed in each hospital ground to cover all study areas. The traps were placed in open areas at about 1.5 meters in height from the soil surface. Also, the insect net was used to collect adult flies.

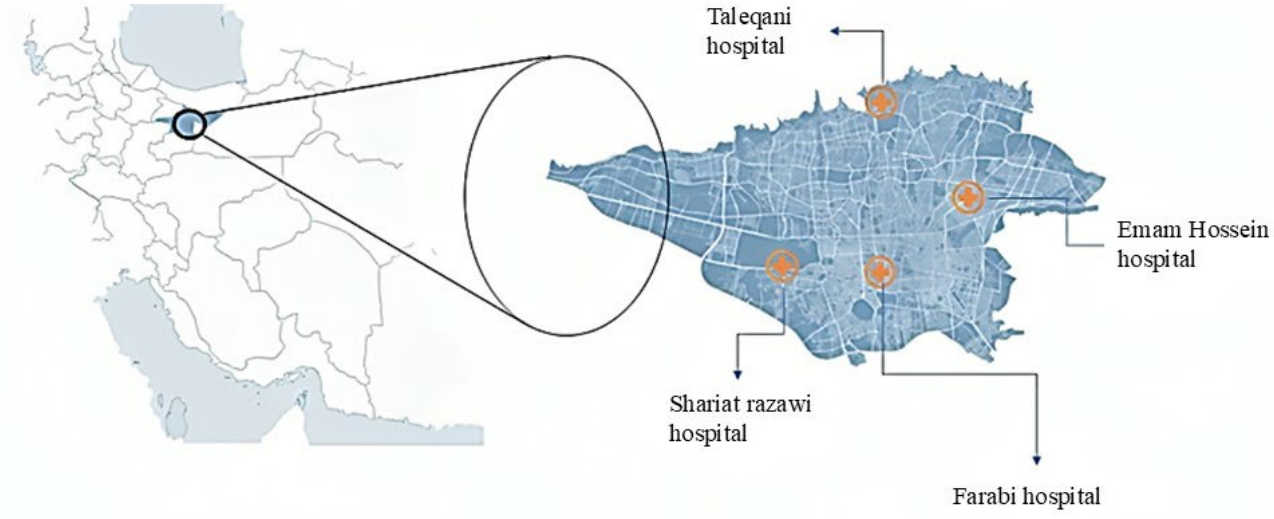

Fig. 1 – The selected locations for sampling, four hospitals in Tehran, Iran.

Fig. 2 – A bottle trap, installed in Taleghani Hospital ground, Tehran, Iran (original photo).

Net collection was done for about one hour in each hospital ground at each collecting interval. A biweekly schedule was approved for sampling from March 2020 to February 2021. The collected adult samples were identified after pinning. The humidifying process was done for the case of dried samples for pulling out the genital organs in some samples such as Sarcophagidae. Some of the collected larvae were reared temporarily to have their adult for confirming their adult fly's identifications. The flies were identified by relevant keys (James 1947; Zumpt 1965; Povolny & Verves 1997; Akbarzadeh et al 2015).

The abundance of any species was calculated by dividing the number of collected samples by eight because of eight hanged bottle traps. All population data for this study have been transformed using square roots calculation to make the data uniform. The results of the square root have been used for statistical analysis. As usual in fly sampling, some saprophagous flies are not caught by bottle traps. In this study, insect nets were used to sample more fly species. The collected samples with insect nets weren't included in the abundance calculations. The raw data have been used for the results and discussion in the text. The transformed abundances were compared based on trapping sites (hospitals) for estimating spatial variations, and seasonal activities for estimating temporal variations, for each species through Kruskal-Wallis test in the SPSS software 19®.

Results

A total of 18613 adult flies have been collected using 32 installed traps of which 8588 (46.14%), 5019 (26.97%), 3045 (16.36%), and 1959 (10.52%) belong to Calliphoridae, Muscidae, Sarcophagidae, and Fanniidae, respectively. All samples were from 18 known species that were potential agents for myiasis including some from Sarcophagidae, Muscidae, Calliphoridae, and Fanniidae families (Table 1).

Calliphoridae

A lot of 7386 from 8588 samples of the family Calliphoridae comprised three species including *Calliphora vicina*, *Lucilia sericata* and *Chrysomya albiceps* with relative abundances of 84.88%, 0.47%, and 4.66%, respectively. Other collected samples of this family (1202 samples) belonged to the genus *Pollenia* spp. Data of this genus didn't enter the abundance calculations due to their incomplete identification. Temporal variation analysis of species of the family Calliphoridae has shown that *C. vicina* has a different seasonal activity. Its peak activity was in the colder months of the year. Both *L. sericata* and *Ch. albiceps* had a peak of activity in mid-summer. Statistical analysis didn't show significant differences among population data of each species during various seasons, $p = 0.87$ for *C. vicina*, $p = 0.65$ for *L. sericata*, and $p = 0.78$ for *Ch. albiceps*. Analysis of spatial distribution showed that members of the family Calliphoridae were uniform across all sampling sites. The *Calliphora vicina* was collected in a dominantly high number. Statistical analysis showed significant differences between the population data of members of Calliphoridae in various hospital environments ($p = 0.001$).

Table 1 - Identified species from the environment of the selected hospitals in Tehran.

Number	Family	Sub family	Genus	Species
1	Sarcophagidae	Paramacronychiinae	*Wohlfahrtia*	*Wohlfahrtia nuba* Wiedemann, 1830
2	"	Miltograminae	*Miltogramma*	*Miltogramma taeniata* Meigen, 1824
3	"	Sarcophaginae	*Sarcophaga*	*Sarcophaga argyrostoma* Robineau-Desvoidy, 1830
4	"	"	"	*S. aegyptica* Salem, 1935
5	"	"	"	*S. africa* Wiedemann, 1824
6	"	"	"	*S. variegata* Scopoli, 1763
7	"	"	"	*S. nigriventris* Meigen, 1826
8	"	"	"	*S. melanura* Meigen, 1826
9	"	"	*Ravinia*	*Ravinia pernix* Harris, 1780
10	Calliphoridae	Calliphorinae	*Calliphora*	*Calliphora vicina* Robineau-Desvoidy, 1830
11	"	Luciliinae	*Lucilia*	*Lucilia sericata* Meigen, 1826
12	"	Chrysomyinae	*Chrysomyia*	*Chrysomyia albiceps* (Wiedemann, 1819)
13	Muscidae	Muscinae	*Musca*	*Musca domestica* Linnaeus, 1758
14	"	Azeliinae	*Muscina*	*Muscina stabulans* Fallen, 1817
15	"	"	"	*Mu. prolapsa* Harris, 1780
16	"	"	"	*Mu. levida* (Harris, 1780)
17	"	"	*Hydrotea*	*Hydrotaea ignava* (Harris, 1780)
18	Fanniidae	--	*Fannia*	*Fannia canicularis* Linnaeus, 1761

Muscidae and Fanniidae

Four species of Muscidae family have been collected in bottle traps in selected hospitals of Tehran including *Muscina stabulans*, *Hydrotaea ignava*, *Muscina prolapsa*, and *Musca domestica* with relative abundances of 73%, 13%, 7%, and 1%, respectively. A few samples of *Mu. levida* have been collected by insect net.

Temporal variations of Muscidae and Fanniidae showed that they are active mostly in moderate and cold weather, spring, autumn, and winter. It seems that, *Mu. stabulans* has the highest activity throughout the year but statistical analysis didn't show a significant difference for the data of this species in various seasons ($p = 0.16$). The other species differ significantly among seasons, $p = 0.03$ for *M. domestica*, $p = 0.01$ for *M. prolapsa*, $p = 0.04$ for *H. ignava,* and $p = 0.03$ for *F. canicularis*.

Except for *H. ignava* ($p = 0.001$) the population data of all the collected species in various hospitals didn't show any significant difference, $p = 0.65$ for *M. domestica*, $p = 0.11$ for *Mu. stabulans*, $p = 0.78$ for *M. prolapsa* and $p = 0.46$ for *F. canicularis*.

Sarcorf

Due to difficulties in the morphological identification of females of Sarcophagidae, about 53% of the samples were left unidentified and mentioned as *Sarcophaga* sp. It was necessary to pull out the genital appendages of male samples of the family Sarcophagidae for their identification. The dominant species of this family was *Sarcophaga africa* which comprised more than 28% of all samples. The other species were *S. aegyptica*, *S. argyrostoma*, and *S. variegata* with relative abundances of 11%, 5%, and 3%, respectively. A few samples of *Wohlfahrtia nuba*, *S. melanura*, *S. nigriventris* (Fig 3), *Miltogramma taeniata,* and *Ravinia pernix* have been found in the study area by net collection. Temporal variations of collected species of Sarcophagidae showed that all of the members of this family are active during warmer months. Statistical analysis revealed that the data of any species varied significantly during various seasons, $p = 0.005$ for *S. argyrostoma*, $p = 0.001$ for *S. aegyptica*, $p = 0.04$ for *S. africa,* and $p = 0.001$ for *S. variegata.* Despite the high collected numbers of species *S. africa*, the statistical analysis didn't show any significant difference between the data of all collected species in various hospital environments ($p = 0.78$).

Discussion

There are gaps in the study of medically important flies in Iran (Akbarzadeh et al 2012). The study on the ability to transmit pathogens by synanthropic flies has been focused mostly on *M. domestica* (Kassiri et al 2012). The study on agents of myiasis has been limited to case reports without more basic studies on their agents (Akbarzadeh et al 2012; Alizadeh et al 2014). Similarly, the study on forensically important flies refers to limited works (Babapour 2015). The results of this study improved the knowledge about the fauna of medically important flies in Tehran and Iran which will be applicable for future works.

The total number of fly species in this study was eighteen 18. This result was similar to the results of Sanei-Dehkordi et al (2020). That study was done in the hospital environments of Bandar Abbas, a port city in the south of Iran, on the northern coastline of the Persian Gulf. The considerable result of that study was the presence of some species including *Chrysomya megacephala* (Fabricius, 1794), *Sarcophaga ruficornis* Fabricius, 1794 and *Sarcophaga*

dux Thomson, 1896 in the south (Sanei-Dehkordi et al 2020). These species didn't report from northern parts of Iran such as Tehran.

The majority of the collected flies in this study belong to the family Calliphoridae, but in the latest work on flies in Fars province, south of Iran (Akbarzadeh 2012), the Muscidae family was the most dominant. This difference may be due to the intervals between the bi-weekly sampling process in this study and monthly sampling in Fars province study (Akbarzadeh 2012). The baits in monthly sampling were more attractive for *M. domestica.*

The dominant species in this study was *C. vicina*, a similar result to Hwang and Turner (2005) in London (Hwang &Turner 2005) and also similar to the fauna of other parts of Iran like Fars (Akbarzadeh 2012), Qom (Mozaffari et al 2020), Sistan and Balochestan (Nateghpour & Akbarzadeh 2017), and some other central parts of Iran. Due to the latest study on the medically important flies of Tehran, *Ch. albiceps* accompanied by *L. sericata* and *M. domestica* have been introduced for cadaver biotope and *M. domestica* for the biotope of garbage and rubbish (Khoobdel 1998).

All reported species of the family Sarcophagidae in this study are new to the fly fauna of Tehran. Khoobdel (1998) reported some species other than the findings of our study. All collected species in this study except for *S. nigriventris*, have been reported from Fars province (Akbarzadeh 2012). *Sarcophaga nigriventris* is being reported for the first time for Iranian fauna (Schumann 1986). A literature review showed one species of Muscidae family, *H. ignava*, being reported for the first time from Tehran. These species have not been reported in the last research in Tehran (Khoobdel 1998).

Fig. 3 – *Sarcophaga nigriventris* Meigen, 1826 (Diptera: Sarcophagidae), collected in hospital grounds of Tehran, Iran (original photo).

The higher abundance of *Mu. stabulans* in comparison with *M. domestica* in Tehran is another new study finding. More than 73% of all samples from the Muscidae family belonged to *Mu. stabulans*. The abundance of *M. domestica* in this research was about 1% of the family, similar to results of the study on medically important flies in Iranian Lesser and Greater and Abomusa Tonb in the Persian Gulf (Khoobdel et al 2013; 2015). In a study in an urban area in Argentina, the higher abundance of *Mu. stabulans* in comparison with *M. domestica* has been confirmed (Patitucci et al 2011). Due to the similarity in ecology and biology of these two species (Service 2012), the study of surface pathogens of *Mu. stabulans* instead of *M. domestica* in hospital environments can be recommended.

The myiasis agents in hospital environments are usually facultative and accidental (Joo & Kim 2001). None of the reported flies in this study are obligatory myiasis. The reported flies in this study can enter the buildings and

produce myiasis. Due to disability of hospitalized patients, low levels of immunization, and available wounds, the producing myiasis may be more dangerous for the victims (Mowlavi et al 2011).

Except for *H. ignava*, the population size of the other species in this study didn't show any significant difference among the hospital environments. This issue proves the overall uniformity in maintenance and management in the studied hospitals. Therefore, some similar controlling measures may be useful for all hospitals in Tehran.

It can be concluded that *C. vicina* can be mentioned as the main species with the potential of producing myiasis, due to its presence everywhere and every time in the study area. The high abundance of *Mu. stabulans* and its similar ecology and biology to *M. domestica* are enough reasons to change the position of this species as the main synanthropic fly in the study area. Moreover, according to the results of this study, the hospital environment could be a suitable place for the breeding of medically important flies. This also emphasizes the importance of fixed nets on the windows and other equipment for entrances to keep the flies far from hospitalized patients.

Author's Contributions

Abbas Ali Mirzakhanlou: investigation, draft preparation; **Mansoureh Shayeghi:** Supervision, visualization; **Hassan Vatandoost:** supervision, project administration; **Arman Izadian:** draft preparation, formal analysis, **Zahra Karimi:** methodology, formal analysis; **Kamran Akbarzadeh:** conceptualization, methodology, project administration, final review and edit

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Funding

Tehran University of Medical Science has financially supported this study with project number 91-04-27-20122.

Data Availability Statement

All data supporting the findings of this study are available within the paper.

Acknowledgments

Tehran University of Medical Science, project number 91-04-27-20122, has financially supported this study. The authors wish to thank some colleagues of Nicolas Copernicus University of Poland for their kind help in the identification of some samples as well as all personnel, especially the security personnel in selected hospitals, for their cooperation in conducting the work.

Ethics Approval

Only insects were used in this study. All applicable international, national, and institutional guidelines for the care and use of animals were followed. This article does not contain any studies with human participants performed by any of the authors.

Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

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Citation: Mirzakhanlou, A. A., Shayeghi, M., Vatandoust, H., Izadian, A., Karimi, Z., and Akbarzadeh, K. (2025) Species diversity of saprophagous flies (Diptera) in hospital grounds of Tehran, Iran. *J. Entomol. Soc. Iran*, 45(1), 97–106.

DOI: https://doi.org/10.61186/jesi.45.1.8

URL: https://jesi.areeo.ac.ir/article_131275.html

pISSN: 0259-9996 – eISSN: 2783-3968

Journal of Entomological Society of Iran

2025, 45 (1), 97–106

تنوع گونه‌ای مگس های پوسیده خوار (Diptera) در محیط های بیمارستانی شهر تهران، ایران

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چکیده: مگس‌های پوسیده خوار از عوامل اساسی انتقال پاتوژن های مختلف و ایجاد میاز بوده و کاربرد آنها در حشره شناسی پزشکی قانونی ضروری است. هدف اصلی این مطالعه شناسایی مگس‌های مهم پزشکی در محوطه بیمارستان‌های تهران بود. فرآیند نمونه‌برداری در این مطالعه مقطعی، هر دو هفته یکبار در محیط‌های ۴ بیمارستان شهر تهران با استفاده از تله‌های بطری و تور حشره‌گیری از اسفند ۱۳۹۹ تا بهمن ۱۴۰۰ انجام شد.به دلیل تغییرات زیاد داده های جمعیتی در طول زمان مورد بررسی، پراکنش داده‌ها با استفاده از محاسبه ریشه دوم، به شکل نرمال تغییر یافتند. داده‌ها با آزمون Kruskal-Wallis برای برآورد تغییرات زمانی و مکانی مورد تجزیه و تحلیل قرار گرفتند. در مجموع ۱۸۶۱۳ مگس بالغ جمع آوری و ۱۸ گونه شناسایی شد. این مگس‌ها به چهار خانواده Calliphoridae، Muscidae، Sarcophagidae و Fanniidae تعلق داشتند. شایع ترین گونه ها در هر خانواده به ترتیب *Calliphora vicina* Robineau-Desvoidy، *Muscina stabulans* Fallen، *Sarcophaga Africa* Wiedemann و *Fannia canicularis* L. بودند. نتایج این مطالعه نشان داد که *C. vicina* فعالیت فصلی متفاوتی دارد، بیشترین وفور آن در ماه‌های سردتر می باشد، در حالی که *Lucilia sericata* و *Chrysomia albiceps* و همچنین گونه غالب خانواده Sarcophagidae در میانه تابستان بیشترین فراوانی آن ها می باشد. تجزیه و تحلیل وفور منطقه‌ای نشان داد تفاوت قابل ملاحظه‌ای از هر سه گونه Calliphoridae در بیمارستان‌ها وجود ندارد. همین امر برای Muscidae و Fanniidae نیز مشاهده شد. گونه غالب در بین گونه‌های خانواده Sarcophagidae در ماههای گرم سال فعال بود. گونه *Sarcophaga nigriventris* Meigen برای اولین بار از ایران گزارش می‌شود.

کلمات کلیدی: میازیس بیمارستانی، حشره شناسی قانونی، تله مگس های بالغ

تاریخچه مقاله

دریافت: ۱۴۰۳/۰۶/۰۸

پذیرش: ۱۴۰۳/۰۹/۲۱

انتشار: ۱۴۰۳/۱۰/۲۱

دبیر تخصصی: علیرضا سازمند

نویسنده مسئول: کامران اکبرزاده

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DOI: https://doi.org/10.61186/jesi.45.1.8