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RESEARCH ARTICLE

IDENTIFICATION OF MEDICALLY-IMPORTANT DIPTERAN SPECIES IN NUWEIBA CITY, SOUTH SINAI, EGYPT, AND THEIR RELATIVE ABUNDANCE

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ABSTRACT

Nuweiba city, South Sinai, Egypt is one of the poorly-studied areas in terms of the presence of medically-important dipteran species. Therefore, in the current study a survey of flies (order: Diptera) of medical importance in Nuweiba city was carried out during winter and summer of 2019. Six traps baited with two sources of attractants – decayed fish pieces and a ready-made botanical extract – were used to collect the flies in four selected localities. A total of 374 individuals belonging to 11 species within 9 genera under 6 families (Culicidae, Ephydridae, Ulidiidae, Calliphoridae, Muscidae, and Sarcophagidae) were captured, and the identification key was provided. The highest numbers of adult flies were found in summer in "Habiba organic farm" using traps baited with decayed fish pieces (n=159). Two species (*Musca domestica* L. and *Culex pipiens* L.) were the most predominant species with a total of 141 and 100 captured specimens, respectively. The collected specimens included medically important species, as disease vectors and myiasis producers in humans and animals, which belong to three families (Calliphoridae, Muscidae, and Sarcophagidae); besides the two mosquito species (*Culex pipiens* and *Culiseta longiareolata* Macquart) that are known as vectors for many diseases of humans. In addition, *Sarcophaga* (*Liopygia*) *surcoufi* is newly recorded in Sinai. In conclusion, the aforementioned dipterous species may have the potential of transmitting diseases to the community dwellers in Nuweiba city; therefore, they must be controlled.

INTRODUCTION

Diptera is among the medically important insects to humans and constitutes more than 120000 described species worldwide[1]. Flies can be mechanical vectors of enteric pathogens including medically important bacteria, and they are also a major vector of important parasites including protozoa and intestinal nematodes or soil-transmitted helminthes[2-4]. Also, they can carry the viruses and fungi to humans[5].

Mosquitoes (Diptera: Culicidae) are important vectors for a wide range of pathogenic organisms and is the deadliest animal in the world. In Egypt, many studies have been carried out to explore the presence

of mosquitoes at several localities including the Red Sea Coast[6], Delta governorates[7], El Sharqiya[8,9], El Qalyobia[10], and El Menoufia[11]. Other studies concerned with the abundance and distribution of calliphorid and sarcophagid species in Nile Valley and Delta, Eastern and Western Deserts, and South Sinai (El Tor) were done[12-15]. However, in these previously mentioned studies Nuweiba city (South Sinai) is one of the areas that have not been monitored so far in terms of the presence of medically important dipterous species. Accordingly, this study is conducted to explore the dipterous insects in Nuweiba city, which have medical importance.

MATERIAL AND METHODS

Experimental sites

This research was carried out in Nuweiba city in 2019 over two seasons, winter (January) and summer (July and September). Nuweiba city (Figure 1) lies on the Northeastern corner of Egypt with about 40 km^2 total area. Nuweiba port was built in 1985 on the Gulf of Aqaba to support the trading and transportation between Egypt, Saudi Arabia, and Jordan. Four experimental localities were selected that represent the regions of the main daily human activity: Habiba beach lodge (29° 1' 20.74" E and 34° 40' 21.20" N), Habiba organic farm (29° 96' 78" E and 34.66° 96' 33" N), and some irrigation reservoirs (29° 96' 5" E and 34.66° 95' 60" N), both drainage canals and opened ponds of the drainage system. Nuweiba port could not be visited due to security reasons. Survey studies were carried out in the previously mentioned selected sites to record different dipterous pests in every season.

Adult fly collection

Adults were collected using traps baited with two types of attractants, decayed fish pieces and a ready-made botanical extract (ELITE Integrated Supplies, Cairo, Egypt), where each was used separately in an insect bait trap. Six traps had been built up in the agriculture habitat and animal sheds in Habiba organic farm in addition to three traps at Habiba beach lodge per each season. Traps were examined every 48 hours to collect the trapped specimens, which were then killed in jars containing tissue papers wetted with few drops of ethyl acetate. The killed specimens were pinned directly to maintain their color and texture. Small-sized specimens were preserved in 70% glyceride-ethanol. Finally, the specimens of each trap catchment were transferred to the laboratory for sorting and identification.

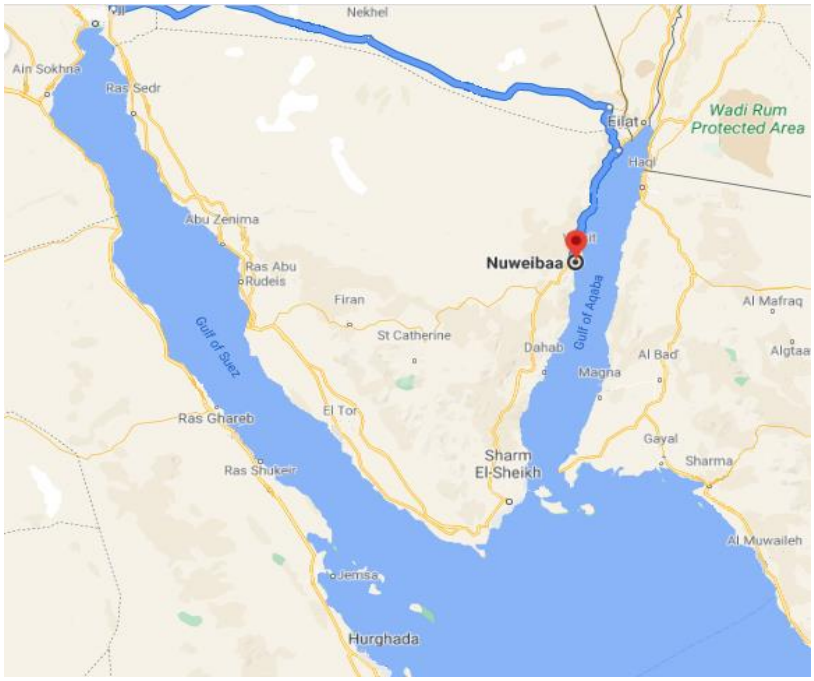

Figure 1: Map of Sinai showing Nuweiba city.

Mosquito larvae collection

Dipper and dropper were used to collect larvae from the drainage ponds and irrigation reservoirs. From these water sources, 24 catches had been seasonally taken. Larvae were collected from the edges of both drainage ponds and irrigation reservoirs at different time intervals during the day except at night. While dipping, samples had been taken from certain collecting stations in the morning to avoid casting shadows on the water surface. Also, samples were collected in the afternoon to avoid shadow from the habitat.

Identification of specimens

Specimens were identified using the valid taxonomic keys and detailed species descripttion[16-30] and names were updated[31]. Images were captured using a Nikon 7100d camera (Minato city, Tokyo, Japan) and microscopic samples were photographed using an S-EYE YW500 camera and Nokia 6 phone camera fixed on a BOECO BM-120 microscope (Hamburg, Germany).

RESULTS

As a result of this study, 259 adult flies belonging to six families, seven genera, and nine species were captured. These included *Musca domestica* (Linnaeus, 1758), *Chlorichaeta tuberculosa* (Becker, 1922), *Physiphora alceae* (Preyssler, 1791), *Wohlfartia nuba* (Wiedemann, 1830), *Sarcophaga surcoufi* (Villeneuve, 1913), *Sarcophaga aegyptiaca* (Salem, 1935), *Chrysomya megacephala* (Fabricius, 1794), *Chrysomya alibiceps* (Wiedemann, 1819) and *Lucilia sericata* (Meigen, 1826) (Tables 1 and 2). In addition, 115 mosquito larvae of the two species, *Culex pipiens* (Linnaeus, 1758) and *Culiseta longiareolata* (Macquart, 1838), were collected (Table 3).

Table (1): Numbers of collected dipteran adult species using traps baited with ready-made botanical extract.

Family	Species	Number of individuals			
		Winter		Summer	
		Habitat (1)	Habitat (2)	Habitat (1)	Habitat (2)
Muscidae	*Musca domestica*	3	7	5	11
Sarcophagidae	*Sarcophaga aegyptiaca*	1	1	1	0
Ulidiidae	*Physiphora alceae*	0	0	15	10
Total number of caught individuals/season		12		42	
Total number of caught individuals		54			

Table (2): Numbers of collected dipteran adult species using traps baited with decayed fish pieces.

Family	Species	Number of individuals			
		Winter		Summer	
		Habitat (1)	Habitat (2)	Habitat (1)	Habitat (2)
Muscidae	*Musca domestica*	7	8	20	80
Ephydridae	*Chlorichaeta tuberculosa*	0	0	0	40
Sarcophagidae	*Sarcophaga aegyptiaca*	1	3	0	5
	Sarcophaga surcoufi	0	0	0	15
	Wohlfartia nuba	0	0	0	15
Calliphoridae	*Chrysomya megacephala*	0	5	0	0
	Chrysomya alibiceps	0	0	0	4
	Lucilia sericata	0	2	0	0
Total number of caught individuals/season		26		179	
Total number of caught individuals		205			

Table (3): Numbers of collected culicid larvae and *Artemia* sp. using dipper and dropper technique.

Species	Number of individuals			
	Winter		Summer	
	Drainage ponds	Irrigation reservoirs	Drainage ponds	Irrigation reservoirs
Culex pipiens	80	20	0	0
Culiseta longiareolata	5	10	0	0
Total number of caught individuals/season	115		0	
Total number of caught individuals	115			
Artemia sp.	~ 200 individuals/100 m. of water in drainage ponds at summer season			

Family Muscidae was the most abundant family (Figure 2) with total of 256 collected specimens during the study. A total of 206 flies, represented nine species were caught from Habiba organic farm (Habitat 2) during the survey period using the two sources of attractants, comparing with 53 flies within three species, *Musca domestica*, *Sarcophaga* (*Lios.*) *aegyptiaca* and *Physiphora alceae* from Habiba beach (Habitat 1) (Figure 3). *Musca domestica* was dominant in the two habitats as 35 and 106 flies were captured, respectively.

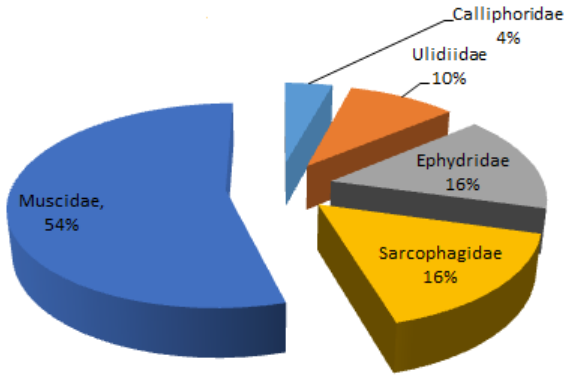

Figure 2: Abundance of dipterous families collected from Nuweiba city in 2019.

Both attractant sources that used in the present study showed variable capturing capability (Figure 4), where the potentiality of decayed fish pieces to attract genera, species, and individuals (6, 8, and 205, respectively) was more than ready-made botanical extract (3, 3, and 54 respectively).

The present results also showed a remarkable increase in the number of individuals, which entrapped during the summer season (221 specimens) compared with those captured at the winter (38 specimens), as shown in Figure "5", with the preponderance of the decayed fish attractant in the two seasons.

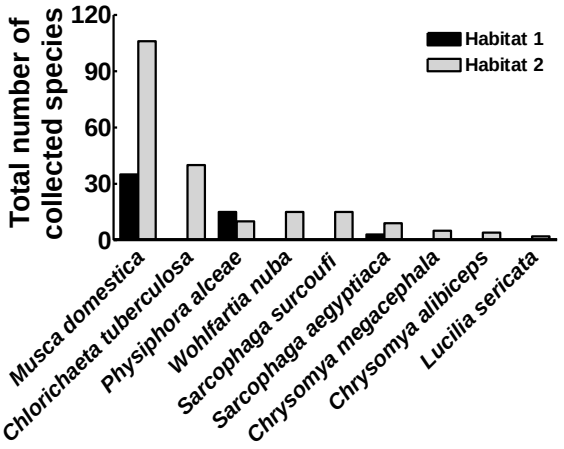

Figure 3: Total number of collected species/habitat.

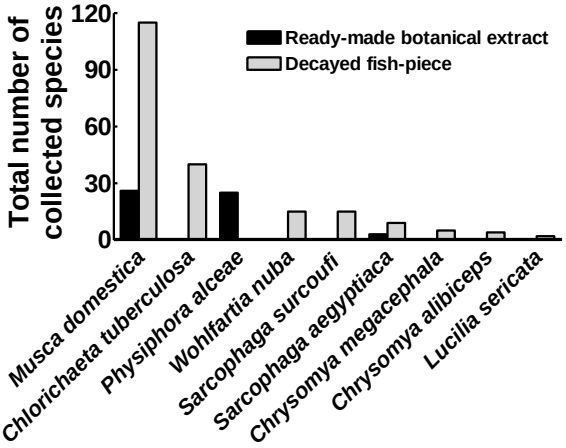

Figure 4: Total number of collected species/attractant source.

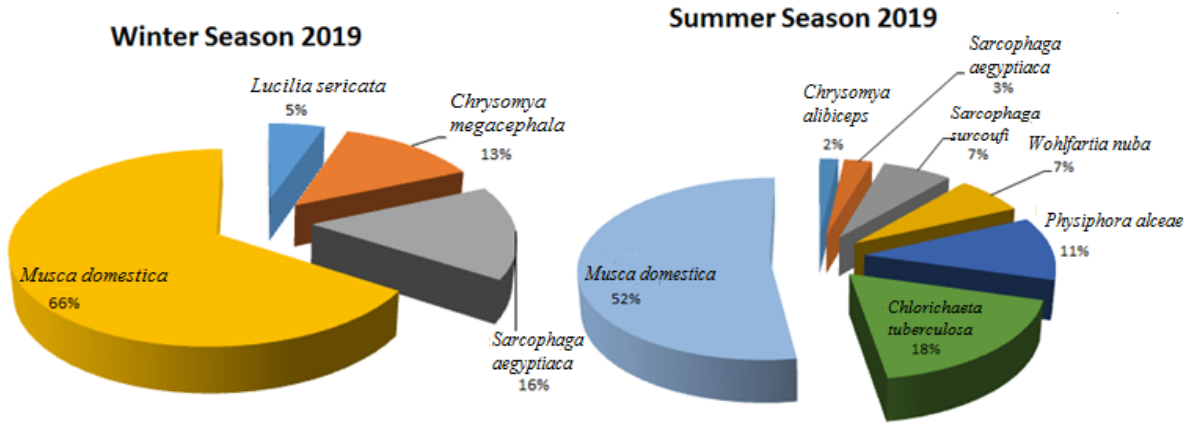

Figure 5: Abundance of dipterous species collected from Nuweiba city in 2019.

In addition, the two mosquito species were caught from the irrigation reservoirs and the drainage ponds only during winter (Figure 6), and no individuals appeared at the summer.

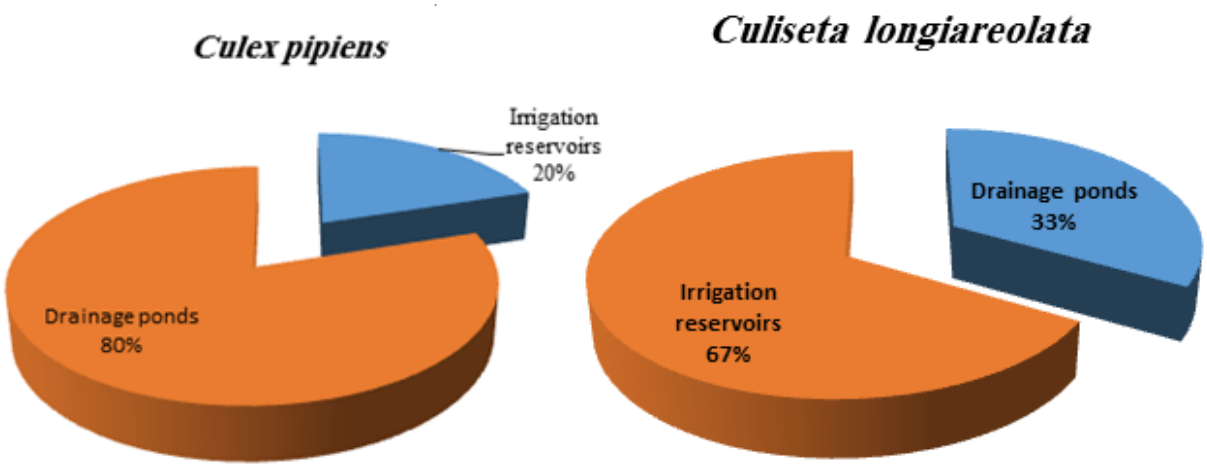

Figure 6: Relative abundance of culicid larval species/collection site.

Key to genera and species of the surveyed 4th instar larvae of family Culicidae from Nuweiba city, South Sinai

1. Siphon with at least three pairs of seta ==➔ *Culex* "Siphon/saddle index 3.45 ==➔ *Culex pipiens* (Figure 7a,b)"
- Siphon with one pair of seta ==➔ *Culiseta* "Pectin spines stout and widely spaced; siphon/saddle index 2.5 ==➔ *Culiseta longiareolata* (Figure 7c,d)"

Key to families of the collected adult flies of medical importance from Nuweiba city, South Sinai

1. Antennal pedicel without dorsal cleft, calypters small or undeveloped ==➔ Acalypetrate ==➔ 2
- Antennal pedicel with a complete dorsal cleft, calypters usually well-developed ==➔ 3
2. Costa is complete, not partially interrupted at or near tip of R1 cup cell (Figure 8f) usually with pointed or extended to posterior apex ==➔ Ulidiidae "Eyes with dark colored stripes, frons reddish brown; wing hyaline, cell r_{4+5} narrowly opened; abdomen black-brown, with purplish blue reflection; male ejaculatory apodeme wide fan like; female ovipositor prominent, with spherical spermathecae ==➔ [*Physiphora alceae* (Preyssler), Figure "8a-j"]"

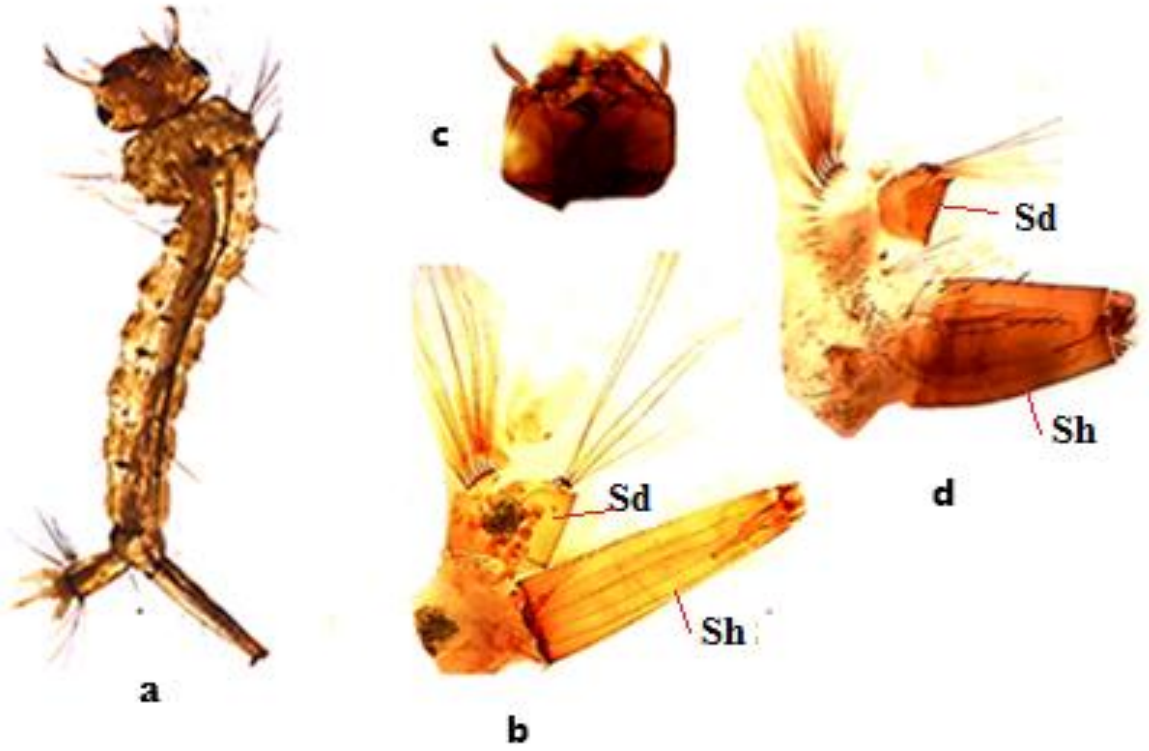

Figure 7: Family Culicidae: (**a** and **b**) *Culex pipiens*, (**a**) larva, (**b**) siphon, (**c** and **d**) *Culiseta longiareolata*, (**c**) head (dorsal view), (**d**) siphon. Sd: saddle, Sh: siphon.

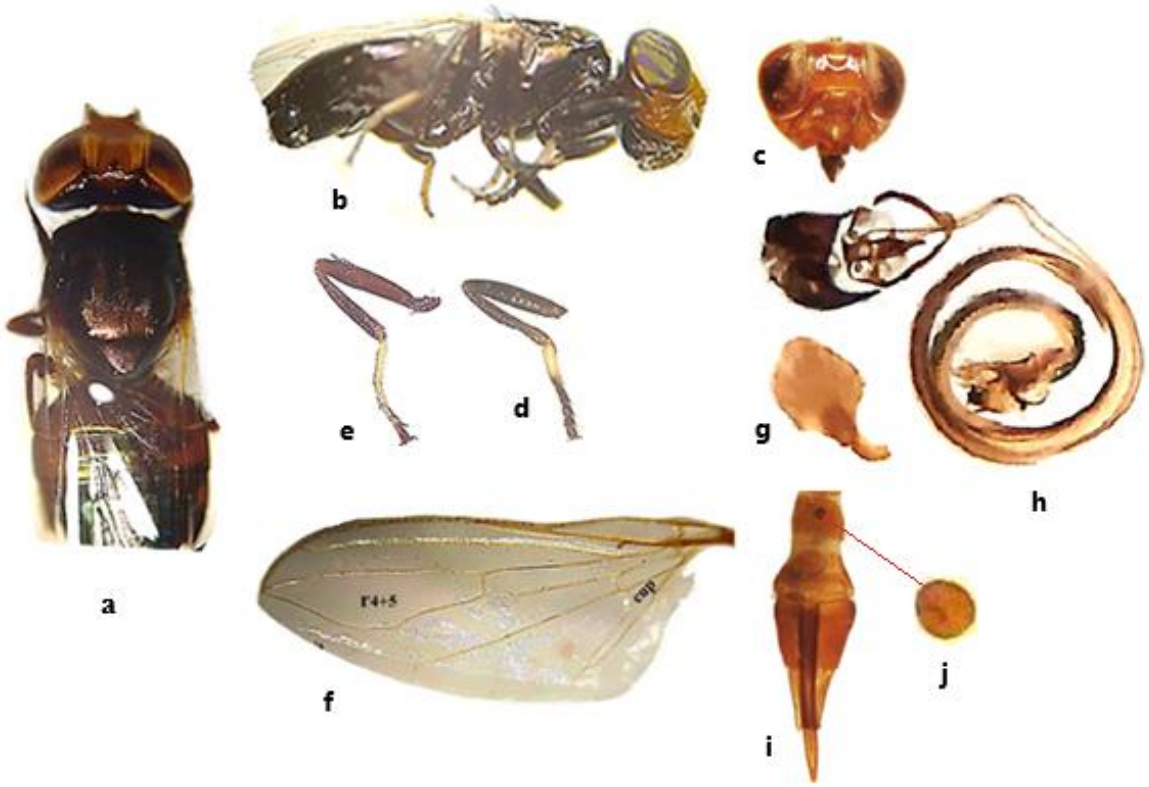

Figure 8: Family Ulidiidae, *Physiphora alceae*: (**a** and **b**) habitus, (**a**) dorsal view, (**b**) lateral view, (**c**) head, frontal view, (**d**) fore leg, (**e**) hind leg, (**f**) fore wing, (**g**) male ejaculatory apodeme, (**h**) male epandrium and phallus, (**i**) female terminalia, (**j**) spermathecal.

- Costa distinctly interrupted before tip of vein R1, (Figure 9e) ==➔ Ephydridae "Head with protruding shiny carinate face; scutellum provided with 6 marginal setae; fore femur with stout spines; length of epandrium + surstyli 1.5 times as width, lateral margins of surstylus concave towards narrowly rounded apex ==➔ [*Chlorichaeta tuberculosa* (Becker), Figure "9a-n"]"

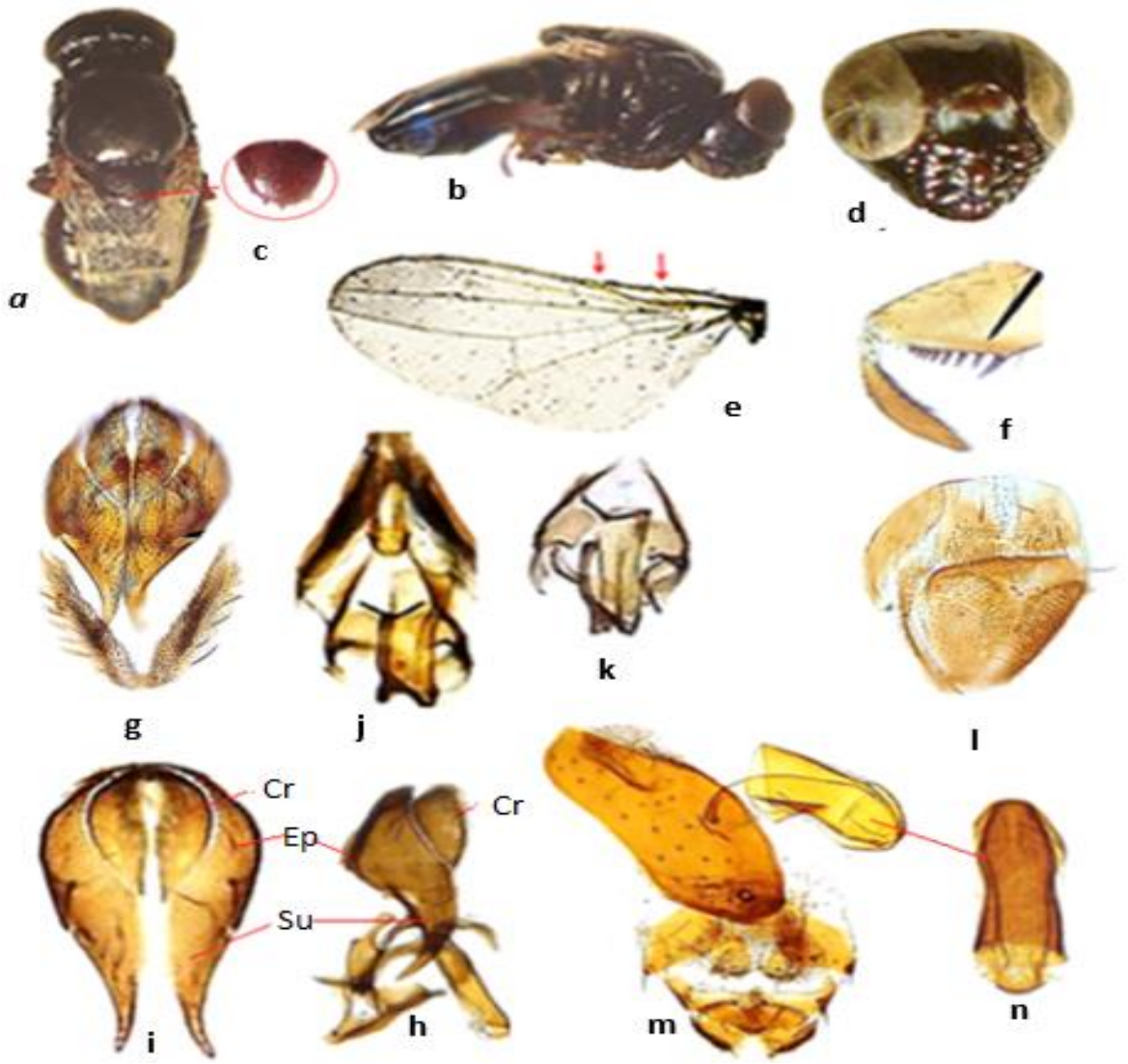

Figure 9: Family Ephydridae, *Chlorichaeta tuberculosa*: (**a** and **b**) habitus, (**a**) dorsal view, (**b**) lateral view, (**c**) scutellum showing marginal setae, (**d**) frontal view of head, (**e**) fore wing rows show interruption in costa (arrows), (**f**) fore femoral spines, (**g-k**) male genital parts, (**g**) dorsal view of male genitalia, (**h**) lateral view of male genitalia, (**i**) epandrium, cerci, and surstyli, (ventral view), (**j** and **k**) aedeagal apodeme and aedeagus, (**l** and **m**): female genital parts, (**n**) female spermatheca. Cr: cercus, Ep: epandrium, Su: surstylus.

3. Meron without a row of setae, rarely with scattered weak setulae ==➔ Muscidae "*Musca domestica* (Linnaeus)"
- Meron with a row of setae (Figure 10a,c) ==➔ 4
4. Abdomen and usually thorax with shining metallic blue, green or bronze colors, notopleura usually with two or three setae ==➔ Calliphoridae ==➔ 5
- Abdomen and thorax dull gray or brown, notopleura usually with four setae (two strong and two small ones) ==➔ Sarcophagidae ==➔ 7

5. Base of stem vein -R bare above, lower calypter bare above ==➔ *Lucilia* "frons at narrowest point 0.12-0.14 as wide of head in male; 0.35-0.40 in female; costa yellow at base; calypters white; supra squamal ridge with a cluster of setae near base of scutellum; posterior spiracles pear-shaped, thorax and abdomen completely metallic, green or bronze ==➔ *Lucilia sericata*"
- Base of stem vein -R hairy above (Figure 11b); squamae hairy ==➔ *Chrysomyia* ==➔ 6
6. Anterior thoracic spiracle pale whitish yellow (Figure 11d), genae totally or for the greater part yellowish, innerside of 3rd antennal segment dark brown; eye facets of male with no clear line of demarcation between larger and smaller facets ==➔ *Chrysomyia albiceps*
- Anterior thoracic spiracle pale brown (Figure 11f), genial dilation with orange hairs, 3rd antennal segment yellowish to orange, eye of male with upper facets much enlarged and with sharp demarcation from small facets below ==➔ *Chrysomyia megacephala*
7. Abdomen with patterns forming a median strip and dark lateral spots, hind coxa bare on posterior surface ==➔ *Wohlfahrtia* "Body medium in size; 5–12 mm in length; gray in color; 2nd antennal segment and palps orange color; thorax gray, with three black stripes; wing with 4th vein sharply angled; abdomen grey with three black round spots at the end of each segment ==➔ *Wohlfahrtia nuba* (Figure 10)"
- Abdomen with distinct check board patterns, hind coxae with setae on posterior surface ==➔ *Sarcophaga* ==➔ 8

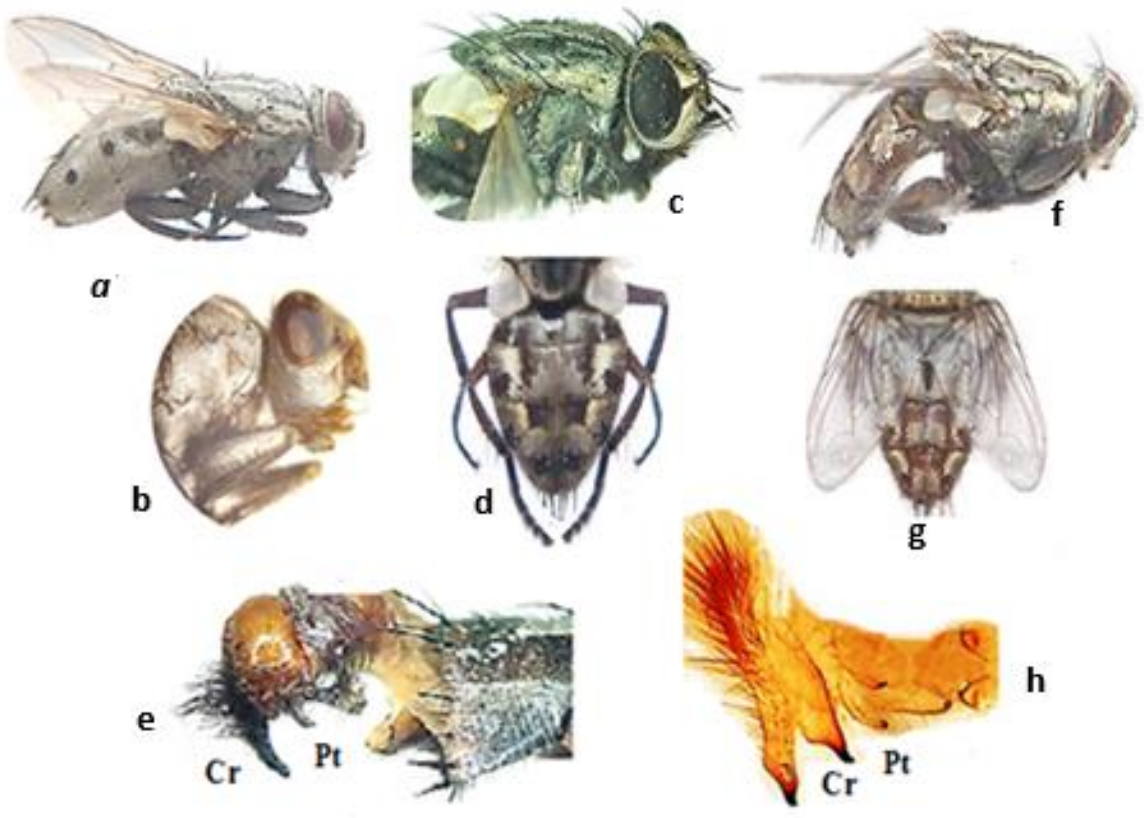

Figure 10: Family Sarcophagidae: (**a** and **b**) *Wohlfartia nuba*, (**a**) habitus (lateral view), (**b**) head (lateral view), (**c-e**) *Sarcophaga aegyptiaca*, (**c**) head and thorax (lateral view), (**d**) abdomen (dorsal view), (**e**) male genitalia (lateral view), (**f-h**) *Sarcophaga* (*Liopygia*) *surcoufi*, (**f**) habitus (lateral view), (**g**) abdomen (dorsal view), (**h**) male genitalia (lateral view). Cr: cercus, Pt: postgonite.

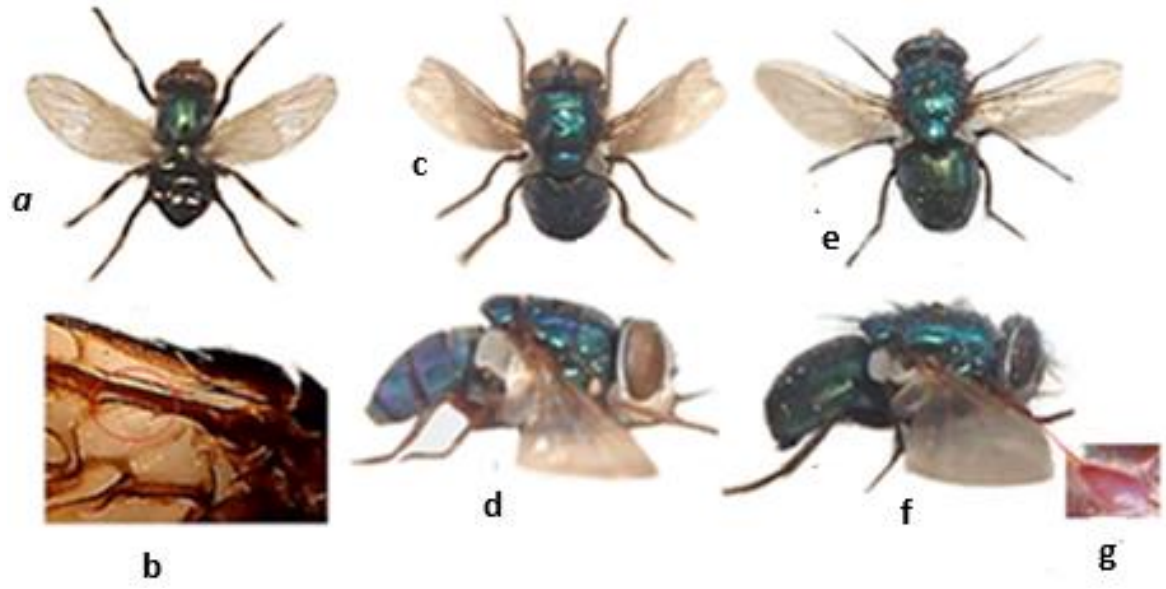

Figure 11: Family Calliphoridae: (**a**) *Lucilia sericata*, habitus (dorsal view), (**b-d**) *Chrysomya alibiceps*, (**b**) base of fore wing, (**c**) habitus (dorsal view), (**d**) habitus (lateral view), (**e-f**) *Chrysomya megacephala*, (**e**) habitus (dorsal view), (**f**) habitus (lateral view), (**g**) anterior spiracle.

8. Gena with white hairs, sometimes with some black hairs at anterior margin as well, Syntergo sternite 7+8 orange, with a grey strip and setose at apical margin; parafacial setae arranged in one row closed to eye ==➔ Subgenus *Liopygia* "Cerci narrow with prominence dorsally, its apical half almost as broad as postgonite; vesica with a long arm ==➔ *S.* (*Liop*) *surcoufi* (Figure 10f-h)"

- Gena with black hairs at anterior half and white hairs at posterior half; Syntergo sternite 7+8 grey at apical half and brown at basal half, without setae at apical margin ==➔ Subgenus *Liosarcophaga* "Epandrium orange; vesica sclerotised, dark, with two short processes apically ==➔ *S.* (*Lios.*) *aegyptica* (Figure 10c-e)"

DISCUSSION

This study represented an updated knowledge to the fauna of the dipteran insects, which have medical and veterinary importance in Nuweiba city (South Sinai) that is considered as a commercial center. According to the results obtained from this research eleven species belonging to nine genera within six families (Culicidae, Ephydridae, Ulidiidae, Calliphoridae, Muscidae, and Sarcophagidae) were collected during the two seasons (summer and winter 2019). The results of the present study revealed that there was a marked difference in the density of the caught medically important flies according to the nature of habitat, attractant sources, and inventory season. On this matter, the present results explain that both attractant sources showed variable capturing ability. This variation in entrapment ability may be due to the different active ingredients that emitted from the two attractants. The effectiveness of the decayed fish pieces source to attract dipterous species of medical importance was also previously mentioned[14]. It is clear that the survey season and the place where the traps got installed may present more suitable harbor for one or some species than others. Among the captured species, only the two species *Musca domestica* and *Sarcophaga* (*Lios.*) *aegyptiaca* were trapped by both attractants, where the composites released from both attractants were successful to attract muscid and sarcophagid species, while the remaining attracted to the fragrances

emitted from only one of the tested attractants.

Regarding the surveyed habitats, the high number of *Musca domestica* that collected from Habiba organic farm may be due to the availability of oviposition and breeding sites, as plant remnants or leftovers in addition to animal droppings or dung. Also, these conditions in Habiba farm may possibly be the main reason for the high abundance of the veterinary important calliphorid and sarcophagid species. In addition, The greater number of specimens, which collected in the summer season, might be due to the faster growth rates and the shortening of the developmental intervals of the immature stages of the flies[32].

The dipper and dropper technique was used to collect and track the seasonal abundance of mosquito larvae. As a result, two mosquito species under two genera belonging to family Culicidae were collected besides *Artemia* sp., brine shrimp (Class: Branchiopoda, Family: Artemiidae). The two mosquito species were caught only during winter this may be due to the appropriateness of the water as an oviposition and breeding sites for the immature stages of mosquito in the winter. The high temperature as well as the high evaporation rate of water bodies in Nuwebia city during the summer may possibly be the main reasons of water unsuitability, and therefore forming the obstacles for the population growth of the mosquitoes. Worthy of note is that no species belonging to genus *Anopheles*, the main vector of malaria, was caught throughout the study. This may be due to the lack of suitable breeding sites/habitat for this mosquito species. Also, *Artemia* sp. was highly dominant in the drainage ponds during summer.

The different abundant patterns of mosquito larvae might be explained by go over the characteristics of their habitat. In this regard, *C. pipiens* were collected from both irrigation reservoirs and drainage ponds that agree with our data[8,11,20,33]. In contrast, the larvae of *Cx. longiareolata* are mainly lived in small fresh water lakes[8]. The finding of *Cx. longiareolata* in drainage ponds might be due to the relative enhancement of the properties of the drainage water by the overflowed water during the flooding season. This running water can refresh the drainage water and dilute the concentration of the sludge or settled it down thus being a suitable habitat for these mosquitos' larvae. The finding of *Artemia* sp. in summer season was of quite different than the case of mosquito larvae. The life behavior of this species might help for explanation. The *Artemia* sp. is adapted to alive in the excessive saline lakes; consequently it is act as a biological indicator for detecting the potential hazardous chemicals in the water samples[34]. The high evaporation rate in the opened drainage ponds of Nuwebia city during summer season may concentrate the salinity of the water, making it a proper habitat for *Artemia* sp.

Regarding the role played by the collected species as disease vectors and myiasis producers, *Wohlfahrtia nuba* was stated as an occasional secondary invader of wounds in camel then can cause severe myiasis. The two species, *Chrysomya albiceps* and *C. megacephala*, are also related to myiasis. Furthermore, the later species act as a potential vector of many diseases owing to its close association with human housing; equally it considered being a potential mechanical vector of fecal pathogens. Also, *Lucilia sericata* causes myiasis in sheep and is a vector for pathogens. *Physiphora alceae* can transmit various disease-causing organisms to food material and can reproduce in human corpses[27,30,35,36]. As well, *Musca domestica* is a mechanical or biological vector of many human pathogens and parasites such as typhoid, cholera, dysentery diseases; also it cause accidental myiasis of intestine and urinogenital *Trachoma* virus, leprosy, tuberculosis, and enteric infections…*etc*[18,37,38]. In addition, both *Culex pipiens* and *Culiseta longiareolata* have been indicated as vectors of several

diseases[39]. In conclusion, since the aforementioned dipterous species have the potential of transmitting disease, it could constitute a health risk to the community dwellers in Nuweiba city and must be controlled.

AUTHORS' CONTRIBUTIONS

SMG, MGS, and IIA conceived and designed research; IIA and AAR collected the specimens; SMG, GMMA, and AAR identified and dissected the collected specimens, writing-original draft; GMMA final editing and formatting. All authors have read and agreed to the published version of the manuscript.

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CONFLICT OF INTEREST

The authors have no potential financial conflict of interest.

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تعيين أنواع ثنائيات الأجنحة ذات الأهمية الطبية في مدينة نويبع، جنوب سيناء، مصر ونسب شيوعها

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مدينة نويبع، جنوب سيناء، مصر هي واحدة من المناطق التي لم تدرس بشكل جيد من حيث رصد ما بها من أنواع الحشرات ثنائيات الأجنحة ذات الأهمية الطبية. لذلك، تم في الدراسة الحالية إجراء مسح لأنواع ثنائيات الأجنحة (رتبة: Diptera) ذات الأهمية الطبية في مدينة نويبع خلال فصلي الشتاء والصيف لعام 2019. وقد تم استخدام ست مصايد طُعم بها مصدرين من المواد الجاذبة لقنص الحشرات اليافعة، وهما قطع أسماك متحللة وخلاصة نباتية جاهزة، في أربعة مواقع. وتم تجميع وتعريف 374 عينة تمثل أحد عشر نوعًا ضمن تسعة أجناس تنتمي إلي ست فصائل هي (Culicidae، Ephydridae، Ulidiidae، Calliphoridae، Muscidae، Sarcophagidae)، كما تم تسجيل مفتاح التعريف. وقد تم جمع أكبر عدد من الحشرات اليافعة صيفًا في "مزرعة حبيبة العضوية"، وذلك باستخدام المصايد المطعمة بقطع سمك متحللة (159 عينة). كما وجِدَ أن الذبابة "*Musca domestica*" والبعوضة "*Culex pipiens*" هما أكثر الأنواع شيوعًا بين الأنواع التي تم جمعها بإجمالي "141 و 100 عينة، على التوالي". وتشمل العينات التي تم جمعها أنواعًا هامة طبيًا، مثل ناقلات الأمراض ومنتج النغف في الإنسان والحيوان، وهي تنتمي إلى ثلاث فصائل (Calliphoridae، Muscidae، Sarcophagidae)؛ إلى جانب نوعي البعوض "*Culex pipiens* و *Culiseta longiareolata*" المعروفين كناقل للعديد من الأمراض التي تصيب الإنسان. وقد أضافت الدراسة النوع "*Sarcophaga (Liopygia) surcoufi*" إلى المجموعة الحشرية في سيناء. وتستنتج الدراسة أنه قد يكون للأنواع المذكورة أعلاه القدرة على نقل الأمراض إلى سكان مدينة نويبع؛ مما يستوجب مكافحتها.