

Species Identification of Forensic Important Blowflies (*Diptera, Calliphoridae*) that occur in Jeddah, Kingdom of Saudi Arabia

Layla A.H. Al-Shareef*

King Abdulaziz University, Kingdom of Saudi Arabia

*Corresponding author: Layla A.H. Al-Shareef, Faculty of Science-AI Faisaliah, King Abdulaziz University, Ministry of Education, Kingdom of Saudi Arabia, Tel: +966564469922; E-mail: Layladr@hotmail.com

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Abstract

An important goal in forensic entomology is the identification of necrophagous fly species that breed directly on carcasses. In kingdom of Saudi Arabia there are rarely studies on carrion breeding blowflies. Using rabbit carcasses as a model for study, a key for the identification of three species of blowflies (*Diptera: Calliphoridae*) collected from deferent habitat (desert, urban, agriculture and coastal) in Jeddah city, west region of the Kingdom of Saudi Arabia was prepared. The blowfly species which found were *Chrysomya megacephala*, *Chrysomya marginalis* and *Chrysomya albiceps*. This study was conducted to create database information on necrophagous flies in the Kingdom of Saudi Arabia geographical region, and it is essential to the progress of forensic entomology in the country.

Keywords: *Chrysomya* species; Blowflies; Kingdom of Saudi Arabia

Introduction

Forensic entomology applies insect evidence to legal problems. Compared to the traditional procedures in Forensic Medicine, insects are accuracy even in advanced stages of decomposition (after four to five days postmortem) when postmortem phenomena such as autolysis and putrefaction occur [1,2]. Flies from the family *Calliphoridae* are the first group that arrive to the cadaver [3,4], and attract by the odor produce during the early stages of decomposition. Since they lay eggs on a carrion within minutes after death, the estimation of the age of the oldest present eggs, larvae or pupae will provide a minimum postmortem interval (mPMI) [4-8]. Insect species associated with carrion and the period of life cycle vary according many factors, one of the most important being the geographic region or bio-geo climatic zone. The bio-geo climatic zone includes the habitat, vegetation, soil type and meteorological conditions of the area [9]. Therefore, knowledge of the insect fauna that are attracted to human cadavers in every geographical region is fundamental for the use of flies as forensic indicators, and databases should be developed [10,11]. This sort of information is obtained from field experiments on carrion decomposition, usually made with animal models [12]. However, in the Kingdom of Saudi Arabia only rarely studies have been conducted on this subject and the blowflies are poorly known, while using information from other bio-geographical areas, with different fauna and environmental conditions, may not provide a sufficient degree of accuracy for using flies in forensic cases [13]. For this purpose the current study aims to identify species and start to create a database for forensic important blowflies which present in the kingdom of Saudi Arabia particularly in Jeddah bio-geo climatic zone.

Materials and Methods

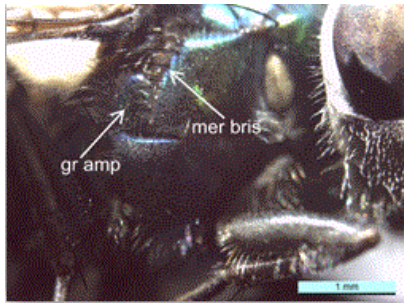
Specimens for this study were obtained from domestic rabbit carcasses located in four deferent habitat (desert, urban, agriculture

and coastal), in Jeddah city in autumn, 2015. Jeddah city is located on the west coast of the Kingdom of Saudi Arabia (latitude 29.21 north & longitude 39.7 east), in the middle of the eastern shore of the Red Sea. The temperature and relative humidity at the time of collecting flies were varied from 26.83°C to 30.62°C and 15% to 89%, respectively. The flies were collected using entomological net and killed in 95% ethyl alcohol, then washed by water with small amount of detergent to remove dust. In some flies real body color was difficult to be observed and seems dull because of the excessively weathered, and we could to solve this problem by placing the specimens in xylene for two hours. The flies were identified with the aid of dissecting stereomicroscope from Leica Company (Leica M205 C stereomicroscope). Digital photographs were taken with Leica IC80 HD camera adapted to a Leica M205 C stereomicroscope. Measurement was given in millimeters. The taxonomy and terminology follows [14-18].

Results

During this study adult fly specimens were obtained from domestic rabbit carcasses located in four deferent habitats (desert, urban, agriculture and coastal) in Jeddah city, kingdom of Saudi Arabia. These flies were observed among the initial colonizers of the corpse, arriving in short time after death and laying their eggs into the natural orifices. All collected flies were belong to family *Calliphoridae* (blowflies), which identified by presence row of bristles on the meron (meral bristles) (Figure 1), absence of prominent subscutellum (Figure 2), third antennal segment has plumose arista (Figure 3), they were medium to large flies (4-16 mm in length) with metallic blue or green thorax and abdomen (Figure 4). All these blowflies from Subfamily; *Chrysomyinae* which easily recognized by being base of stem-vein (R) with row of bristles on the dorsal surface (Figure 5). The flies belong to genus *Chrysomya* Robineau-Desvoidy which characterized by reniform greater ampulla with stiff erect hairs (Figure 1), dorsal surface of lower calypter with dense hairs (Figure 6), dorsum of first abdominal tergite (syntergite 1+2) and posterior margins of tergites 3

and 4 black (Figure 7), genal dilation (cheek) whitish or yellowish (Figures 8- 10).



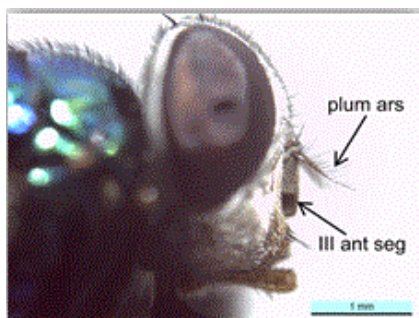
gr amp: greater ampulla
mer bris: meral bristles

Figure 1: Lateral view of *Ch. albiceps* thorax showing row of bristles on the meron and greater ampulla.



scut: scutellum

Figure 2: Lateral dorsal view of *Ch. megacephala* showing absence of prominent subscutellum.



plum ars: plumose arista
III ant seg: third antennal segment

Figure 3: Lateral view of *Ch. albiceps* head showing plumose arista.

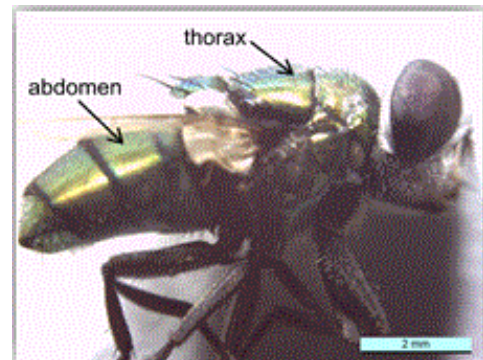
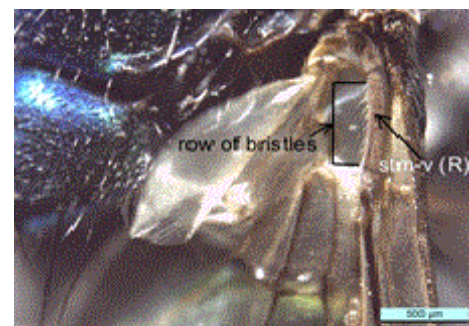
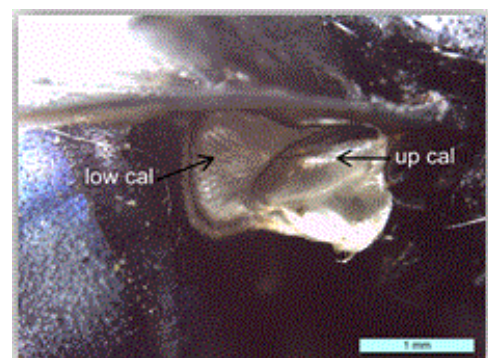


Figure 4: *Ch. albiceps*, shining thorax and abdomen.



stm-v (R): stem-vein (R)

Figure 5: Dorsal view of basal part of wing stem-vein (R) in *Ch. albiceps*.



up cal: upper calypter
low cal: lower calypter

Figure 6: Lateral view of thorax in *Ch. marginalis* showing upper, and lower calypters.

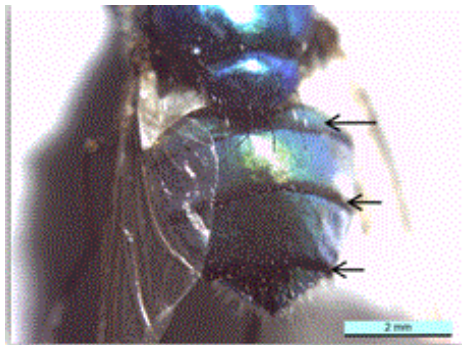
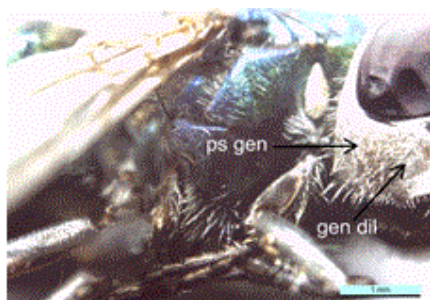
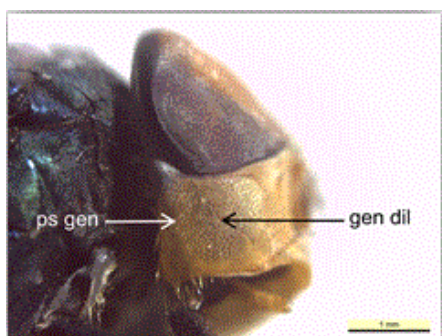


Figure 7: Dorsal view of *Ch. albiceps* abdomen, rows point to black color of first abdominal tergite and posterior margins of tergites 3 and 4.



gen dil: genal dilation
ps gen: post gena

Figure 8: Lateral anterior view of *Ch. albiceps* showing whitish genal dilation.



gen dil: genal dilation
ps gen: post-gena

Figure 9: Lateral anterior view of *Ch. megacephala* showing genal dilation and post-gena.

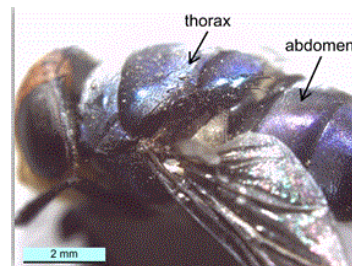


Figure 10: *Ch. marginalis*, metallic dark blue thorax and abdomen.

Key for the identification of the species of Calliphoridae with forensic importance in Jeddah city, Kingdom of Saudi Arabia

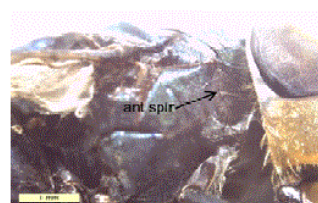
Blowflies from family *Calliphoridae*, subfamily *Chrysominae*, genus *Chrysomya* - 1

1. Anterior spiracle dark- brownish (Figure 11) - *Chrysomya megacephala*

1'. Anterior spiracle white (Figure 12) - 2

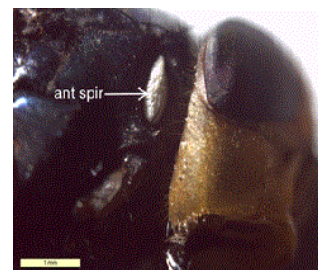
2. Anterior wing margin strongly darkened (Figure 13) - *Chrysomya marginalis*

2'. Anterior wing margin hyaline (Figure 14) - *Chrysomya albiceps*



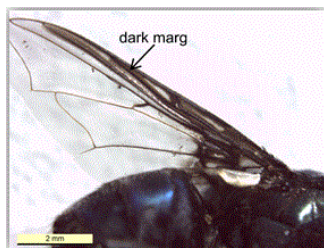
ant spir: anterior spiracle

Figure 11: Lateral anterior view of *Ch. megacephala* showing dark-brownish anterior spiracle.



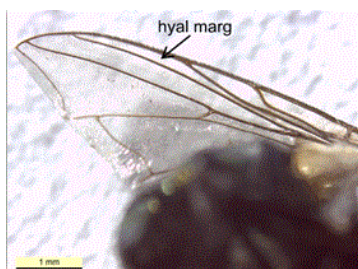
ant spir: anterior spiracle

Figure 12: Lateral anterior view of *Ch. marginalis* showing white anterior spiracle.



dark marg: dark margin

Figure 13: Ventral view of *Ch. marginalis* showing darkened anterior wing margin.

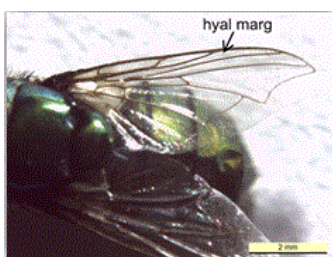


hyal marg: hyaline margin

Figure 14: Ventral view of *Ch. albiceps* showing hyaline anterior wing margin.

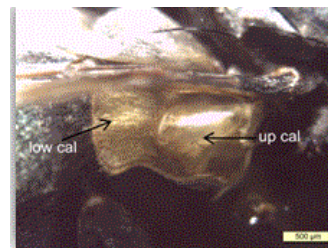
Notes on species

Chrysomya megacephala (Fabricius, 1794): **Diagnosis-** Flies with medium size 6-9 mm in length. Surface of genal dilation and post-gena orange with orange to yellow hair, anterior spiracle brownish (Figure 9). Wings entirely hyaline (Figure 15). Lower calypter yellowish to dirty gray (Figure 16). Male frons very narrow, eyes touching with ommatidia in upper two thirds enlarged and sharply demarcated from small ones in lower third (Figure 17). Frons of female bulging in the middle, not parallel-sided, with dark brown to black color, the fronto-orbital plate and parafacial dark grayish (Figure 18).



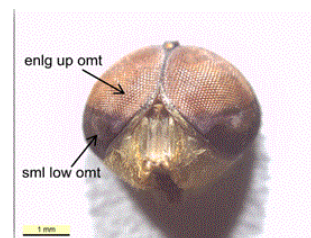
hyal marg: hyaline margin

Figure 15: Dorsal view of *Ch. megacephala*.



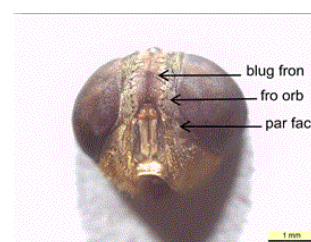
up cal: upper calypter
low cal: lower calypter

Figure 16: Lateral view of *Ch. megacephala* thorax showing yellowish lower calypter.



enlg up omt: enlarged upper ommatidia
sml low omt: small lower ommatidia

Figure 17: Anterior view of male *Ch. megacephala* head showing eyes touching with sharply demarcated and enlarged upper ommatidia.

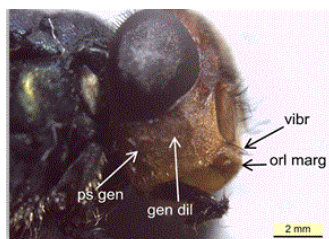


blug fron: bulging frons
fr-orb: fronto-orbital plate
par fac: parafacial

Figure 18: Anterior view of female *Ch. megacephala* head showing dark brown bulging frons, dark-grayish fronto-orbital plate and parafacial.

Distribution: *Chrysomya megacephala* was first recorded in the late 1970's [19-21], then spread quickly and now present in most continents [22]. This species has been recorded in continental Europe [23,24], Mediterranean region [24,25], Palearctic region (Middle East, Canary Is.) [26,27], presently covering the Siberian sub-region, Spain, Iran, Afghanistan, China, Japan, [28], United States [29-34], West Indies, and Brazil [35]. It is also recorded in southern Africa [36], Egypt, Kuwait, Oman, Pakistan, United Arab Emirates, Kingdom of Saudi Arabia [37].

Chrysomya marginalis (Wiedemann, 1819): Diagnosis - Large species, usually 8-12 mm in length. The body is metallic dark blue (Figure 10). Surface of genal dilation and post-gena orange with orange to yellow hairs, vibrissae well above the oral margin (Figure 19). It is characteristic by deeply darkened of anterior margin wings (Figure 11).



vibr: vibrissae ; **orl marg:** oral margin;
gen dil: genal dilation; **ps gen:** post-gena

Figure 19: Lateral anterior view of *Ch. marginalis* showing well developed vibrissae.

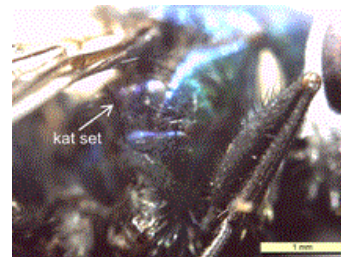
Distribution: This species generally common in sub-Saharan Africa [38]. It was recorded in Burkina Faso, Central African Republic, Egypt, Congo, Kenya, Madagascar, Mali, Namibia, Nigeria, Rwanda, South Africa, Senegal, Somalia, Sudan, Tanzania, Zimbabwe [38-46]. Also occurs in kingdom of Saudi Arabia, Yemen, Oman, United Arab Emirates, Iran, Pakistan [37].

Chrysomya albiceps: Diagnosis - Flies with medium size (6-9 mm in length). Thorax and abdomen metallic blue to green (Figure 4), wings entirely hyaline (Figure 12). Anterior spiracle whitish, proepisternal seta (blew anterior spiracle) absent (Figure 20). Katepisternal setae 1+1 (Figure 21). Male and female vertex with well-developed outer vertical setae (Figure 22). Surface of genal dilation and post-gena whitish with white hairs (Figure 8). Male frons very narrow, eyes very close to each other with ommatidia uniform in size (Figure 23). Frons of female with dark brown to black color, the fronto-orbital plate and parafacial whitish (Figure 24). Third antennal segment wholly dark-grayish (Figure 25). At least some hairs on lateral surfaces of tergite V white, posterior edge of tergite V of female with crevice/incision (Figure 26).



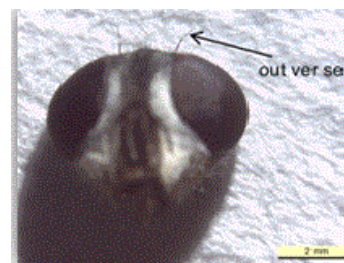
ant spir: anterior spiracle
p ep: proepisternal seta

Figure 20: Lateral view of *Ch. albiceps* thorax showing absence proepisternal seta.



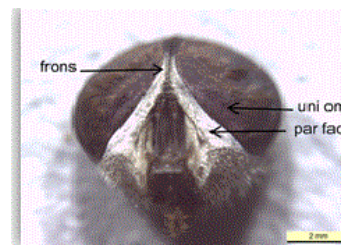
kat set: katepisternal seta

Figure 21: Lateral view of *Ch. albiceps* thorax showing katepisternal setae.



out ver set: outer vertical seta

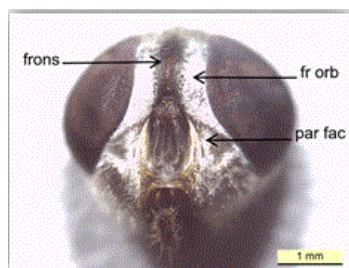
Figure 22: Anterior view of *Ch. albiceps* head showing outer vertical setae.



uni omt: uniform ommatidia
par fac: parafacial

Figure 23: Anterior view of *Ch. albiceps* male head showing eyes with ommatidia uniform in size.

Distribution: *Chrysomya albiceps* is very common species in the Mediterranean regions, and has been reported in Egypt, Iran, Iraq, Kuwait, Lebanon, Libya, Oman, Pakistan, Kingdom of Saudi Arabia, Syria, United Arab Emirates and Turkey [37]. It is recently shown expansion towards central Europe [47-49]. This species is already established in Portugal [50] and other European countries [51], recently having reached Poland [49]. It was also recorded in America [16], West Indies.



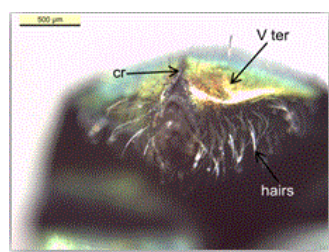
fr orb: fronto-orbital plate
par fac: parafacial

Figure 24: Anterior view of female *Ch. albiceps* head showing dark brown frons, whitish fronto-orbital plate and parafacial.



iii ant seg: third antennal segment

Figure 25: Dark-grayish third antennal segment.



V ter: fifth tergite
cr: crevice/incision

Figure 26: Dorsal view posterior part of female abdomen.

Discussion

Blowflies occur in all biogeography regions and contain around 1500 species [52]. The fauna of blowflies has been studied all over the world; in South America [53], Argentina [54], North America [16,18], Portugal [55], East and South Asia [56], Africa [57]. It was also studied in European and Mediterranean regions [58], Afrotropical region [17]. Blowfly species in the Middle East region were studied by Akbarzadeh et al. [37] and a key was accomplished. However, the fauna of blowflies of forensic importance was not studied enough in the Arabian Gulf region, and there is not any key to identify species in the Kingdom of Saudi Arabia. This situation stops broad application of insects for medico-legal purposes due to a lack of knowledge proper tools for

species identification of the local fauna. According to [53], the definition of the carrion flies faces two major problems: the lack of taxonomists and the lack of keys. Therefore, recent study is a part of large ongoing effort to identify carrion-feeding entomofauna and to start to build a checklist of necrophagous species present in this geographical area; the Kingdom of Saudi Arabia.

The recent study proved that there are three species of blowflies attracted to the rabbit carcasses in Jeddah city, all of them were belong to genus *Chrysomya* Robineau-Desvoidy. Until now, *Chrysomya* Robineau-Desvoidy flies were confined to the Old World tropics and subtropics regions, where they were among the most abundant and economically important blowflies [59].

In this study, *Chrysomya megacephala* (Fabricius) was observed among the first wave of flies visiting rabbit carcasses. This species known as the 'oriental latrine fly' due to its favorite place of ovipositing in latrines [60], and it is may be origins in New Guinea [61]. Larvae are commonly breed in feces and carrion [30,62]. Due to its close association with humans, it was considered to be a potential mechanical vector of faecal pathogens [63]. *Chrysomya megacephala* was recorded in Kuwait by Al-Mesbah et al. [64]. It was used in forensic entomology cases, for postmortem interval determination [65-67]. In the recent study, the blowfly *Chrysomya marginalis* (Wiedemann) was attracted to the rabbit carcasses. This species is normally feces and carrion breeder and one of the primary flies associated with decomposing corpses in areas in which the species is present [53]. It is rarely enters houses, but are common around butcher shops, and also feed from sores and wounds of cattle [62]. Braack [68] found large numbers of *Ch. marginalis* on freshly death impala in Kruger Park. In the Arabian Gulf region *Chrysomya marginalis* was reported in the Kingdom of Saudi Arabia on rabbit carcasses in the mountains of Al-Baha Province at southwestern area [69]. It was also recorded in United Arab Emirates, Oman and Yemen [37]. One of the most abundant blowflies species associated with rabbit carcasses in this study was *Chrysomya albiceps*. It is strong fliers, and the maximum dispersal rates for this species were estimated as 16 km/12 days [70]. *Chrysomya albiceps* larvae are predatory on other larvae present in carrion [62]. They are typically found in carrion but are also able to consume decomposing flesh on wounds of living animals, and considered a primary myiasis fly [26]. Adults prefer high temperatures and humidity, and can be attracted in large numbers by meat [62]. Buttiker et al. [71] proved that *Chrysomya albiceps* is endophilic in Saudi Arabia and a nuisance in homes and markets there.

Conclusion

It was recorded in the Kingdom of Saudi Arabia by [69] in Al-Baha Province at southwestern region, and in Jeddah city at the west region by [72], it was also reported in Qatar [73] and in Kuwait [64]. Al-Ghamdi et al. [74] collected *Chrysomya megacephala* and *Chrysomya albiceps* from different locations of Jeddah city, and they mentioned some morphological characters for these two species which were not enough to understand taxonomical features, also they didn't prepare any taxonomical key.

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