Isolation and Identification of Intestinal parasites and protozoa from Flesh flies *Sarcophaga africa* in Baghdad

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**Abstract**
A study was conducted to determine the role of flesh flies *Sarcophaga africa* as carriers of intestinal parasites in different locations of Baghdad as a primary efforts in Iraq from March to October 2011. A total of 140 *Sarcophaga africa* were examined both the external and internal to identify the parasites. The parasites were transmitted externally: (*Paramphistomum* sp. 10.7%, *Ascaris vitulorum* 7.8%, *Strongyloides westri* 5%, *Taenia* sp. 10.6%). The parasites were transmitted internally: (*Trichuris* sp. 2.8%, *Toxocara* sp. 20% and *Ascaris lubrecoides* 12.8%, *Cryptosporidium parvum* 17.8%, *Cryptosporidium muris* 8.5%, *Cyclospora cayetanensis* 6.8%, *Thieleria* sp. 5% and *Trypanosoma* sp. 2.1%). It was observed that more parasites were isolated from gut contents than the external surfaces of the flies examined (P ≤ 0.01). The public health significance of these findings is highlighted.

**Keywords:** *Sarcophaga Africa*, intestinal parasites, protozoa, mechanical transmission, flesh fly.

**Introduction**
Flesh flies are common, conspicuous flies. This insect measures about 12mm from head to tail. Flesh flies differ from the tachinid flies in that they lack the postscutellum, the large swelling underneath the scutellum on the thorax, [1]. Most flesh flies breed in carrion, dung, or decaying material, but a few species lay their eggs in the open wounds of mammals; hence their common name, [2]. Some flesh fly larvae are internal parasites of other insects, some of them have been reported as parasites on snails, turtles and tortoises, [3,4]. The *Sarcophaga* species is noted for its dark red, forward-facing eyes. It is found almost worldwide except South America, [5].

*Sarcophaga africa* is a species of fly belonging to the family Sarcophagidae, the flesh-flies. It is the best known species in its genus, [6]. *S. africa* feeds on living and dead tissue, including snails, and other decomposing matter, and feces, [7]. *S. africa* is a synanthropic species known to cause Myiasis in humans and livestock, [8]. The species is considered useful in forensic entomology due to this phenomena, [9]. *S. africa* is coprophagous lays eggs in feces, and can be cultured from human and animal feces, [6]. The fly also lays eggs in decaying flesh and can be cultured from the decaying matter, [6]. The aim of this survey is to revealed the role of flesh fly *Sarcophaga africa* as carriers of pathogens as a primary effort of Iraq.

**Material and Methods**
A total of 1123 different flies were collected from slaughter markets of ruminants and fishes in Baghdad. In addition, traps were put in zoo and slaughter of equine in Al-Zawra Park. The traps are perforated metal box and aerial nets. The baits were Silurid fish. The box was put at 50cm. above the land. A total of 140 fly were chosen and diagnosed as *Sarcophaga africa* by Natural History Researches center and museum; from March to October 2011, so as to identify the eggs, larvae and cysts of protozoa.

Two methods applied:
1- Washing technique: all flies were put down in distal water for 3 hours and centrifuge the supernatant to identify the parasites that transmitted mechanically, [10].
2- Staining technique: The procedure that was used with gut contents of flesh fly after deposit the abdomen; to identify the eggs, larvae and cyst of protozoa that transmitted internally by two stains technique: Zeal Nelson stain (80) flies and Giemsa stain (60) flies, [11].

Color Photographs of eggs, larvae and cysts were taken after ocular micrometer calibration [8]. Diagnosis was carried out at Iraqi Natural History Researches center and Museum, University of Baghdad. Key of diagnosis were according to [12,13,14,15].
Results

Washing technique: two species of nematode eggs, (*Ascaris vitulorum* and *Strongyloides westri*), one species of trematode egg (*Paramphistomum* sp.) and one species of cestode egg (*Taenia* sp.) were reported in this study that transmitted mechanically by *Sarcophaga africa* Table (1), Figs. (1), (2), (3), (4).

Where as the Zael Nelson stain technique was showed different figures of Rhabidatae larvae Fig.(5). Four species of protozoa (*Cryptosporidium parvum*, *Cryptosporidium muris*, *Cyclospora cayetanensis* and *Trypanosoma* sp.) from tested of 80 *Sarcophaga africa* fly after stained with Zael Nelson, Table (2), Figs. (6), (7), (8), (9), (10), (11). On the other hand Giemsa stain technique showed two species of nematode eggs, (Ascaris lumbriciodes and *Toxocara* sp.) and one species of protozoa (*Thieleria* sp.) that isolated from 60 *Sarcophaga africa* fly after stained with Giemsa, Table (3), Figs. (12), (13), (14).

| **Table (1)** Numbers of parasites transmitted mechanically by *Sarcophaga africa*. |
|---------------------------------|-----------------|-----------------|-----------------|
| **Species of parasites**       | **Class**       | **No. of parasites** | **%**          |
| *Paramphistomum* sp. egg       | Trematode       | 15               | 10.7           |
| *Ascaris vitulorum* egg        | Nematode        | 11               | 7.8            |
| *Strongyloides westri* egg     | Nematode        | 7                | 5.0            |
| *Taenia* sp. egg               | Cestode         | 6                | 4.2            |

Significant differences P ≤ 0.01 between two methods.

| **Table (2)** Numbers of parasites transmitted by gut contents of *Sarcophaga africa* stained with zeal nelson stain. |
|---------------------------------|-----------------|-----------------|-----------------|
| **Species of parasites**       | **Class**       | **No. of parasites** | **%**          |
| Rhabidatae larvae              | Nematode        | 30               | 21.4           |
| *Crypt. Parvum* cyst           | Protozoa        | 25               | 17.8           |
| *Crypt. Muris* cyst            | Protozoa        | 12               | 8.5            |
| *Cyclospora cayetanensis*      | Protozoa        | 9                | 6.4            |
| *Taenia* sp. egg               | Cestode         | 9                | 6.4            |
| *Trichuris* sp. egg            | Nematode        | 4                | 2.8            |
| *Trypanosoma* sp.              | protozoa        | 3                | 2.1            |

Significant differences P ≤ 0.01 among the species of parasites.

| **Table (3)** Numbers of parasites transmitted by gut contents of *Sarcophaga africa* stained with Giemsa stain. |
|---------------------------------|-----------------|-----------------|-----------------|
| **Species of parasites**       | **Class**       | **No. of parasites** | **%**          |
| *Toxocara* sp. egg             | Nematode        | 28               | 20.0           |
| *Ascaris lubrecoides* Unfertilized egg | Nematode | 18               | 12.8           |
| *Thieleria* sp. ring form      | protozoa        | 7                | 5.0            |
Fig.(1) Paramphistomum sp. egg externally transmitted by flesh fly.

Fig.(2) Ascaris vitulorum egg externally transmitted by flesh fly.

Fig.(3) Strongyloides wastri egg externally transmitted by flesh fly.
Fig. (4) Taenia sp. egg externally transmitted by flesh fly.

Fig. (5) Rhabidatae larvae, internally transmitted by flesh fly.
Fig.(6) Cryptosporidium parvum cyst, internally transmitted by flesh fly.

Fig.(7) Cryptosporidium muris cyst, internally transmitted by flesh fly.

Fig.(8) Cyclospora cayetanensis cyst, internally transmitted by flesh fly.
Fig. (9) *Taenia* sp. egg, internally transmitted by flesh fly.

Fig. (10) *Trichuris* sp. egg, internally transmitted by flesh fly.

Fig. (11) *Trypanosoma* sp. internally transmitted by flesh fly.
Fig.(12) Toxocara sp. egg, internally transmitted by flesh fly.

Fig.(13) Ascaris lumbricoides unfertilized egg, internally transmitted by flesh fly.

Fig.(14) Theileria sp. ring form, internally transmitted by flesh fly.
Discussion

The role of some adult flesh flies *Sarcophaga africa* as carriers of helminthes parasites of man and animals was studied at Baghdad as a primary efforts in Iraq. Eggs of helminthes can be carried both externally and internally by flies. Adherence of eggs to flies externally was shown by Nicoll [16] and Sychevskaya, [17]. This study reviewed four species of parasite eggs transmitted by external surfaces of flesh fly *Sarcophaga africa*: *Paramphistomum* sp., *Ascaris vitulorum*, *Strogyloides westri* and *Taenia* sp. Eggs of up to 0.09 mm can be carried in this way but remain there for only a few hours. The viability and infectivity of such eggs were not discussed but it is unlikely that they are affected: helminth eggs are quite resistant to changes in their external natural environment, [18].

In Baghdad a similar survey of *Musca demostica* showed three species of protozoa (*Entamoeba coli*, *Entamoeba histolytica* and *Iodamoeba* sp.) and four species of nematode (*Ascaris lumbricoides*, *Ascaridia* sp., *Strongyloides* sp. and *Habronema* sp.) carried mechanically by flies, [19].

Internal carriage of eggs by flies was demonstrated in most of the experiments but only Nicoll [16], showed them directly in the intestines by microscopic examination of the intestinal tract. The other experiments were done in an indirect way by examining the deposit of crushed flies or by examining the regurgitations and fecal specks of the flies. The indirect way of showing the internal carriage of eggs of helminthes leaves doubt: body, legs, regurgitations and fecal specks can easily be contaminated by externally-adhering eggs, [17].

The size of the helminthes egg is an important limiting factor for ingestion by flies. Only eggs smaller than 0.05mm can be ingested. Such eggs can remain in the intestinal tract for up to 20 hours, [20].

The rate of Rabditiae larvae was 30(21.4%), this percent apparently is infective in *Sarcophaga africa*, a fly which often breeds in feces and browses on human and animal skin, could have transmission potential, so that *Taenia* sp., *Tichuris* sp. and *Ascaris lumbricoides* were showed in this survey (Table (2)).

*Taenia* sp. appeared in external and internal test that may because this egg was small 30-36µ, easily carried and ingested. Eggs can be carried both externally and internally by flies, size is an impotent limiting factor for ingestion by flies; eggs remain infectious when carried by flies [18], that similar to [21] investigated the flesh fly *Sarcophaga libialia* as transmitted of *Echinococcus granulosus* eggs in human foods.

It was observed that more parasites were isolated from gut contents than the external surfaces of the flies that examined (P ≤ 0.001), that similar to [22].

Protozoan parasites can pass through the fly gastrointestinal tract without alteration of their infectivity and can be subsequently deposited on visited surfaces in "fecal spots". [23]. This study showed five species of protozoa were carried internally by *Sarcophaga africa*. Three of them (*Cryptosporidium parvum*, *Cryptosporidium muris*, and *Cyclospora cayetanensis*) carried by *Musca demostica* documented in previous survey in Baghdad [19]. The prevalence of these parasites in Baghdad in human: *Cryptosporidium* sp.15.15%, *Cyclospora* sp.1.5%.

In rats: *Crypto.sp.* 50% and *Cyclo. Sp.*2.63%, in cats &dogs: *Crypto. Sp.*20%, in vegetables: *Crypto. Sp.*7.40% and *Cyclo. Sp.* 3.70% [11]. *Trypanosoma* sp. and *Thieleria* sp. are protozoan parasites of blood for different animals like ruminants and equine. *Sarcophaga africa* lay their eggs in open wounds of mammals; hence their common name (flesh fly). So appeared of these protozoa in gut contents of flesh fly was accidently.

Biodiversity of these parasites (nematode, cestode, trematode and protozoa P ≤ 0.01) were carried by *Sarcophaga africa* externally and internally made this fly is risk for public health that must be prevent from enter houses, restaurants, hospitals, butcher shops...etc.
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References
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الخلاصة

أهتمت الدراسة بدور ذبابة اللحم نوع Sarcophaga في نقل الطفيليات والأوالي المعوية للمرة من اذار الى نهاية تشرين الأول 2011 من مواقع مختلفة في بغداد، وكجهود اولية في العراق وقد ظهر من فحص 241 ذبابة لحم خارجيا وداخللي الطفيليات المختلفة. الطفيليات الاتية تنقل خارجياً:

(Paramphistomum sp.10.7%, Ascaris vitulorum 7.8%, Strongyloides westri 5%, Taenia sp. 10.6%).

والطفيليات التي تنتقل داخليا كالاتي:

(Trichuris sp.2.8%, Toxocara sp.20% and Ascaris luberecoides12.8%, Cryptosporidium parvum 17.8%,Cryptosporidium muris 8.5%, Cyclospora cayetanensis 6.8%, Thieleria sp. 5% and Trypanosoma sp. 2.1%)

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