

INSIGHTS INTO THE USE OF NATURAL PRODUCTS IN CONTROL OF TICKS AND FLEAS INFESTING DOGS IN EGYPT

By

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ABSTRACT

Ticks and fleas are considered the most common external parasites infesting dogs. Total of (117) dogs were examined for external parasitic infestation; ticks and fleas were collected and subjected for identification. Fecal samples also collected and examined for presence of any internal parasites. Ticks were identified as *R. sanguineus* and fleas as *C. felis*. Epidemiological data (age, sex, breed and season) were recorded. Clinical signs were reported in form of emaciation, anemia, pale mucous membrane, itching, pruritus and alopecia. In vitro and in vivo studies were conducted on parasidose shampoo as a natural therapeutic product for control of ticks and fleas with active ingredient *Biococidine*. It was proved that parasidose effective on both of them and this was the first data recorded from Egypt. Natural oils were used and evaluated as repellent for these external parasites.

Key words:

Ticks, Fleas, Epidemiology, Parasidose, Natural Oils.

INTRODUCTION

The skin is the largest and most visible organ of the body; it is the anatomic and physiologic barrier between animal and environment. It provides protection from physical, chemical and microbiologic injury, and its sensory components perceive heat, cold, pain, pruritus, touch and pressure (Scott *et al.* 1995). The most common canine skin disorders are infestation with ectoparasites. They have a worldwide distribution and are capable of disease transmission. Also cause life-threatening anemia and occasionally hypersensitivity disorders in young and debilitated animals (Araujo *et al.* 1998). Some ectoparasites of pet animals, notably fleas, can infest humans and may lead to the development of dermatitis and transmit vector-borne diseases (Scott *et al.* 2001). Ticks are the second most important arthropods that may transmit

pathogens to dogs (**Franc and Cadiergues 1999**). The economic impact of their treatments is high; it has been estimated globally that pet owners spend more than 2 billion USD annually on flea products alone (**Kramer and Mencke 2001**). Due to the ability of ticks to survive under different climatic conditions and ecological niches, these ticks have a cosmopolitan distribution and their control is still a major challenge for veterinarians and pet owners. The main line of defense against ticks and fleas is the use of chemicals (**Bensignor et al. 2014**). Although chemicals have been effective in suppressing tick and flea populations and incidences of their borne diseases, their main disadvantages have been the high costs, development of acaricide resistance, environmental and food contamination by acaricides and their residues (**Combescot-Lang et al. 2015**). So the scientific community has spurred an interest in developing alternative methods to their control (**Bensignor et al. 2014**). Some natural products like Parasidose shampoo was evaluated for its pediculicidal and ovicidal activity on lice and nits and it was found that it has a physical action on lice. This product consists of Biococidine, Conservators, perfume, and excipients (**Combescot-Lang et al. 2015**). Most plant essential oils are chemically complex, which enhances their efficacy owing to synergy among constituents as recently demonstrated (**Isman et al. 2011**).

MATERIAL AND METHODS

Animals:

In the present study, total number of (117) dogs was examined with great consideration to animal welfare and under owner's permission during the period from November 2014 to November 2017, at the small animal-medicine teaching hospital, faculty of Veterinary Medicine, Cairo University, Egypt. Information about ages, sexes, breeds, season, and clinical signs were recorded.

Tick samples:

Tick samples were collected, by using forceps (anticlockwise). Samples were kept in 70% ethanol for identification according to (**Nuttall and Warburton 1908**).

Flea samples:

Fleas were collected via combing or brushing according to (**Omudu et al. 2010**). Samples were kept in 70% ethanol for identification. All arthropods were identified microscopical at 40X to species identification using the diagnostic keys.

Fecal samples:

Obtained directly from the rectum of healthy and diseased dogs and examined for presence of gastro-intestinal parasites **Farag (2016)**.

In vitro therapeutic study (Heukelbach et al. 2006).

R. sanguineus ticks were collected from infested dogs and were grouped into 3 groups, each group consists of (15 male and 15 female) and tested in vitro in petri dish, the first group immersed in water, the second immersed in Deltamethrin and the third immersed in Parasidose shampoo. *C. felis* were collected and grouped into 2 groups, each group containing 20 fleas and tested in vitro in eppendorf where the first group immersed in water, the second immersed in Parasidose shampoo. Immersion was done for 3 min. and vitality was evaluated for more than 1 h by examination under the dissecting microscope.

In vivo therapeutic study (Franc and Cadiergues 1999).

Sixteen dog were randomly allocated into 2 groups, the first was used as control, and the second was experimentally infested with ticks (*R. sanguineus*) and fleas (*C. felis*). Each group encloses 8 dogs ranging from 2 months - 3 years of age. Experimentally infested dogs were treated with parasidose shampoo (Bottle containing 200 ml shampoo sufficient for 2 months old dog), which applied to the entire coat of the dog, the hair must be dry, left for 45 minutes then the dog's coat was combed thoroughly with special fine toothed stainless steel comb to remove the parasites on a white paper. Then microscopical examinations applied to these parasites to examine the vitality.

Natural oils treatment:

Mixture of essential oils (Citronella oil - Rosemary oil- Peppermint oil - Lavender- pumpkin oil- Camphor oil - Cinnamon oil) were used as a repellent for ticks and fleas after treatment with parasidose shampoo and was applied once every 2-3 days.

Statistical analysis:

Statistical analysis was performed using ANOVA test. Differences were considered significant at $P \leq 0.05$ levels (**Freeman and Tukey 1950**).

RESULTS

During examination of 514 clinical cases admitted to the clinics, the prevalence of tick and flea infestations was in 117 cases of them; Percentage of infestations of dogs by ticks and fleas was 22.7 % of total examined animals; the percentage of tick infestations (67.5%) was higher than flea infestations (19.7%) then mixed infestations (12.8%). The examined dogs was divided according to age into 3 groups, the highest infection rate was recording in dogs less than 1 year with percentage of 74.7 % of ticks infestations, 65.2 % in fleas infestations and 66.7 % in mixed infestations followed by dogs 1 - ≤ 3 years old 21.5 %, 26.1 % and 33.3% then >3 years old 3.8 % and 8.7 % respectively as shown in (Table 1), Fig. (1). this data revealed that, the small aged dogs are more prone to be affected with ticks and fleas infestations than other age groups. Regarding to dog's sex, Percentage of flea and mixed infestations were higher in male (52.2 %),(53.3 %) than female (47.8 %),(46.7 %), but tick infestations were higher in female (55.7 %) than male (44.3 %) as shown in (Table 2), Fig.(2). Concerning to breeds the result showed that, the highest prevalence level was recorded in German Shepherd dogs as ticks, fleas and mixed infestations 54.4 %, 13.04 % and 40 %, followed by Rottweil 18.9%, 21.7% and 26.7% then American Pitbull 10.1%, 30.4% and 26.7% and the least prevalence level was recorded in Great Dane (1.3 % tick infestation), it may be attributed to rarely of this breed in Egypt, as shown in (Table 3) , Fig.(3). Among Season, Percentage of dogs infested with ticks, fleas and mixed infection of both were higher in (spring 59.3%, 80.9%, 76.6% and Summer 35.7%, 19.1 %, 17.6 %) than in (winter 5%, zero, 5.8% and Autumn (Zero) as shown in (Table 4) , Fig, (4).

Table (1): Prevalence of tick and flea infestation among different-age groups.

Groups	Age (yrs)	% of ticks infestations	% of fleas infestations	% of mixed infestations
G1	< 1	74.7	65.2	66.7
G2	1 ≤ 3	21.5	26.1	33.3
G3	>3	3.8	8.7	0
Total % of infested dogs		67.5	19.7	12.8
Total No. of infested dogs	117 (100%)			
Total No. of examined dogs	514			
% of infestations	22.7 %			

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Table (2): Prevalence of dermatological affections among males and females.

Sex	% of ticks infestations	% of fleas infestations	% of mixed infestations
Male	44.3	52.2	53.3
Female	55.7	47.8	46.7
Total % of infested dogs	67.5	19.7	12.8
Total No. of infested dogs	117 (100%)		
Total No. of examined dogs	514		
% of infestations	22.7 %		

Table (3): Prevalence of dermatological problems among different dog breeds.

Breeds	% of ticks infestations	% of fleas infestations	% of mixed infestations
German Shephard	54.4 %	13.04%	40%
Rottweiler	18.9%	21.7%	26.7%
American Pitbull	10.1%	30.4%	26.7 %
Griffone	3.8%	13.04%	0
Labrador Retriever	3.8%	0	0
Golden Retriever	5.1%	8.7%	6.7%
Bullmastive	0	0	0
Cocker Spaniel	0	8.7%	0
Pekingese	0	4.3%	0
Dalmation	2.5 %	0	0
Great Dane	1.3%	0	0
Total No. of infested dogs	117 (100%)		
Total No. of examined dogs	514		

Table (4): Prevalence of dermatological affections among different seasons.

Season	% of ticks infestations	% of fleas infestations	% of mixed infestations
Winter	5	0	5.8
Spring	59.3	80.9	76.6
Summer	35.7	19.1	17.6
Autumn	0	0	0
Total % of infested dogs	67.5	19.7	12.8
Total No. of infested dogs	117 (100%)		
Total No. of examined dogs	514		
% of infestations	22.7 %		

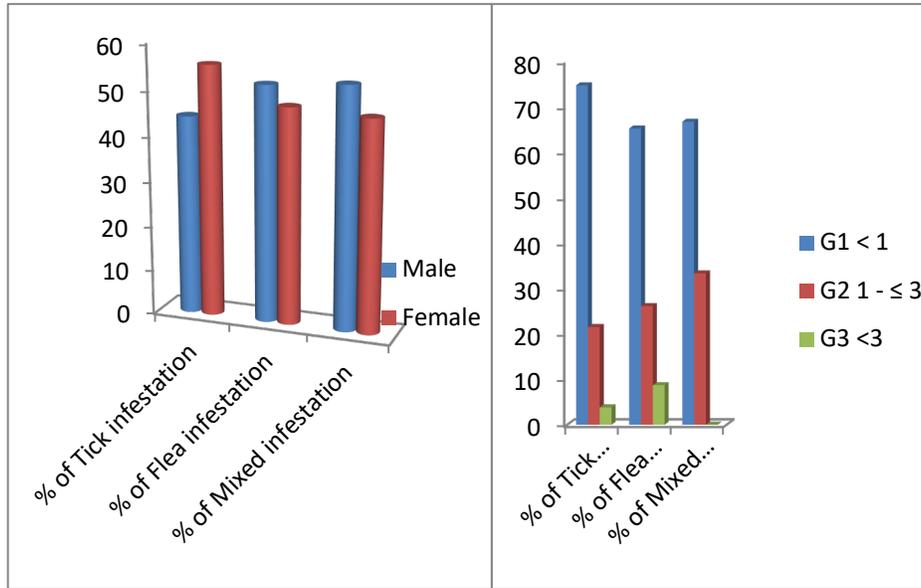


Fig. (1): Prevalence of dermatological affections among different age groups.

Fig. (2): Prevalence of dermatological affections according to sex.

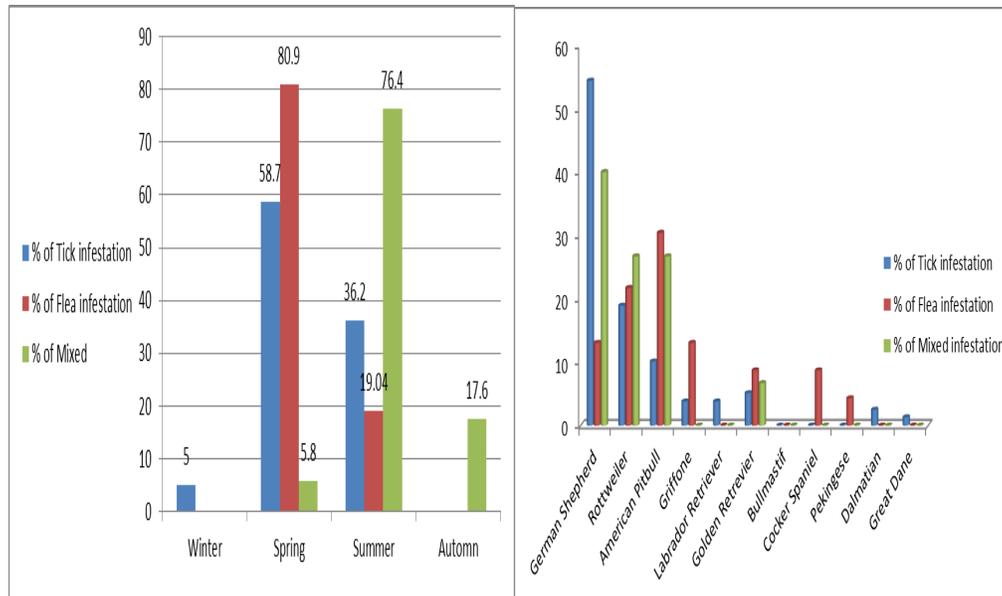


Fig. (3): prevalence of ticks and fleas infestations among breeds.

Fig. (4): prevalence of ticks and fleas infestations among seasons.

The most common clinical signs recorded in dogs infested by ticks and fleas was emaciation, anaemia and pale mucous membrane due to blood loss, itching especially in ear in case of tick infestation, pruritus, alopecic due to severe itching, In flea infestation itching and biting of tail and lumbosacral region, Flea allergic dermatitis (FAD), also found flea excreta when hairs

were dispersed and flea itself as shown in Photo (2). The most common sites for tick infestations was inside ear , between paws, outside ear, head, back , abdomen and belly as shown in photo (1) and for flea infestation was tail, lumbosacral region and belly. Microscopical examination of fecal sample of dogs infested by ectoparasites reveal presence of *Dipylidium caninum* egg nest and *Toxocara canis* adult and egg in some cases.



Photo (1): Tick infestation on different body parts in dogs.



Photo (2): Flea dirt's attached to hair of dogs.

Regarding to in vitro effect of Parasidose shampoo on ticks males, females , eggs and larvae with compared with Deltamethrin and water (control) revealed that mean mortality of male after 15 minutes control 8.89 ± 5.9 , Deltamethrin 91.87 ± 3.85 and parasidose 90.44 ± 5.88 , mean mortality of female after 15 minutes control 10.56 ± 5.87 , Deltamethrin 86.67 ± 3.84 and parasidose 85.78 ± 5.83 , mean of hatchability %control 79.33 ± 2.25 , Deltamethrin 8.33 ± 4.4 and parasidose 10.07 ± 5.23 , mean mortality of larvae after 15 minutes control 0.53 ± 0.03 ,

Deltamethrin 93.87 ± 2.6 , Parasidose 93.47 ± 2.8 , as shown in (Table5) ,Fig.(5). which indicated that there was a significant between parasidose and control, between Deltamethrin and control and no significant between parasidose and Deltamethrin. This was the first data of a field study recorded from Egypt indicated that parasidose shampoo is highly effective in vitro against eggs, larvae, males and females of *R. sanguineus* ticks and fleas, as shown in photo (3-5).

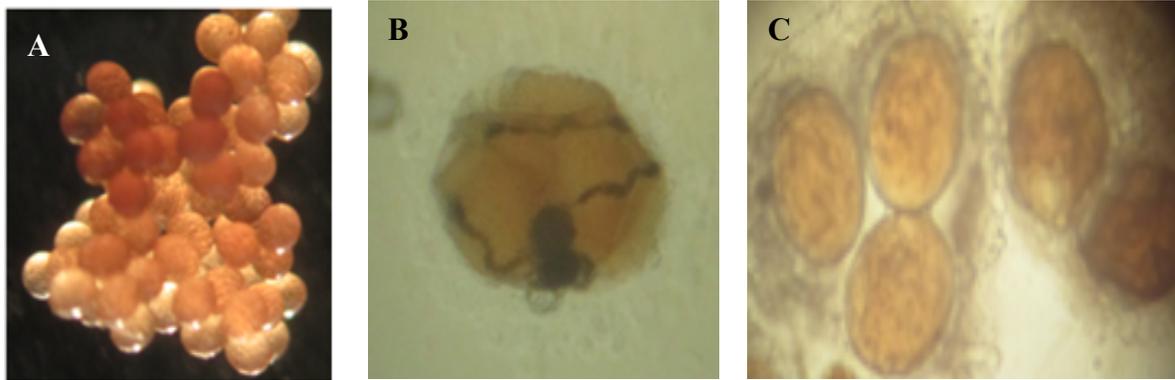


Photo (3): Show eggs of *R. Sanguineus* tick (A) control, (B) and (C) treated by parasidose shampoo.

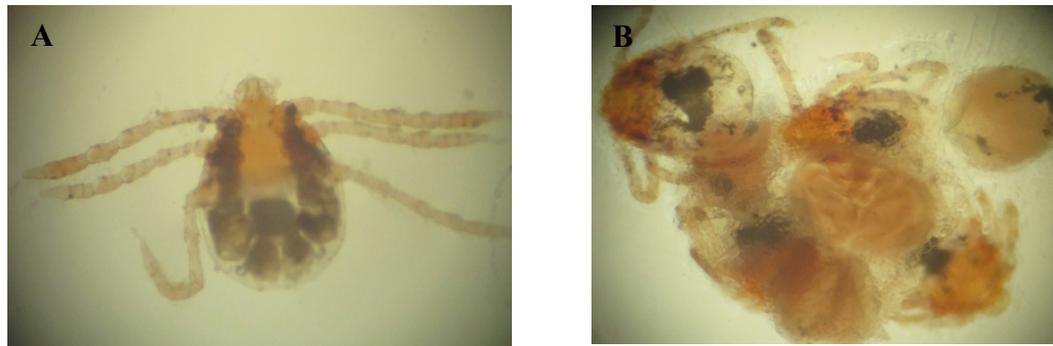


Photo (4): show *R. Sanguineus* tick (A) larva Control and (B) larvae treated by parasidose shampoo.

Table (5): Effect of Deltamethrin and Parasidose on ticks (Mean±SE).

	Control (Water)	Butox (Deltamethrin)	Parasidose (Biococidine)
% of males mortality after 15 min	8.89±5.9^a	91.87±3.85^b	90.44±5.88^b
% of female mortality after 15 min	10.56±5.87^a	86.67±3.84^b	85.78± 5.83^b
% of hatchability	79.33±2.25^b	8.33± 4.4^a	10.07± 5.23^a
% of larvae mortality after 15 min	0.53± 0.03^a	93.87± 2.6^b	93.47± 2.8^b

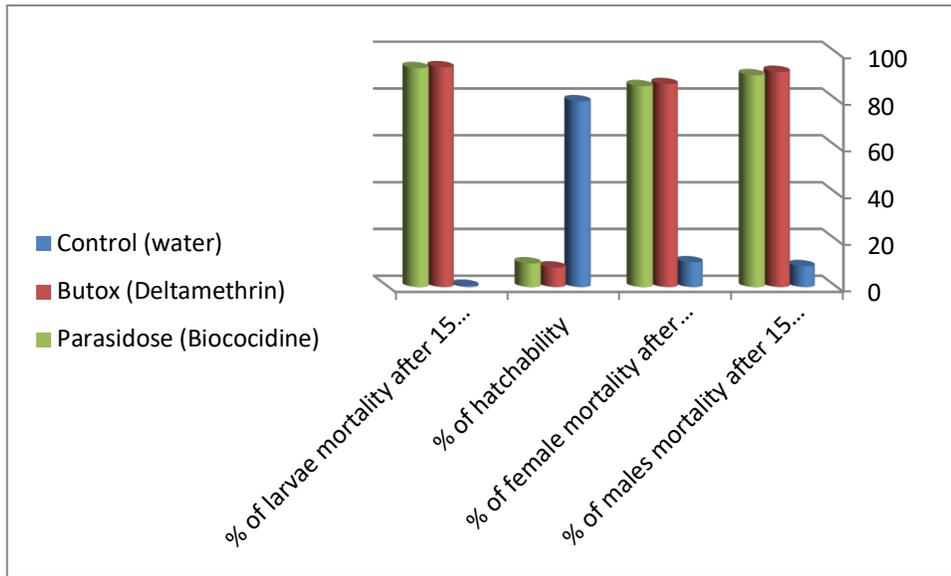


Fig. (5): Show Significant between Parasidose and control and non-significant between parasidose and Deltamethrin (Mean±SE).



Photo (5): Show effect of parasidose on fleas in vitro.

To test in vivo effect of parasidose shampoo. Control and treated group was infested by ticks and fleas. % of attached ticks on pitbull treated was 17.33 ± 5.38 , Rottweiler treated was 47.5 ± 14.8 , pitbull control was 41.67 ± 7.26 and Rottweiler control was 53.5 ± 12.07 . % of dead ticks in treated group pitbull and Rottweiler was 82.65 ± 3.4 and 86.0 ± 2.8 while in control group was 0 ± 0 for both. % of dead ticks after retreatment (after 1 hour) was 100 ± 0.0 for both pitbull and Rottweiler treated while 0 ± 0 for both control group as shown in (Table 6). Regarding to fleas infested dogs, % of attached fleas on pitbull treated was 47 ± 2.38 , Rottweiler treated was 54 ± 14.7 , and pitbull control was 56 ± 11.69 and Rottweiler control was 55.5 ± 10.8 . % of dead fleas in treated group pitbull and Rottweiler was $94.46 \pm 0.2.2$ and 98.55 ± 0.84 while in control group was 0 ± 0 for both. % of dead fleas after retreatment (after 1 hour) was 100 ± 0.0 for both pitbull and Rottweiler treated while 0 ± 0 for both control group, as shown in (Table 7). Which mean there was a significant value between dogs infested with ticks and fleas treated by parasidose shampoo and control group, $P < 0.05$, those was the first data recorded from Egypt indicated that parasidose shampoo is highly effective against ticks and fleas infested dogs as shown in photo (6). Concerning using of mixture of natural oils (Citronella oil-Rosemary oil- Pipermint oil- Lavender- pumpkin oil- Camphor oil - Cinnamon oil) as a repellent of ticks and fleas, dogs were painted by mixture of oils once every 2-3days for repellent of ectoparasites away of it. Repellent effect of ticks (reinfestations after (Days) was 2.95 ± 0.5 to treated bitbull and 2.6 ± 0.58 to treated Rottweiler and 0 ± 0 for control group and for fleas repellent effect (reinfestations after (Days) was 2.75 ± 0.56 to treated Pitbull and 2.577 ± 0.577 to treated Rottweiler and 0 ± 0 for control group. Which mean there was a significant between treated group and control, $P < 0.05$. as shown in (Table 6 , 7).

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Photo (6): Show dogs infested by ticks (A) before and (B) after treatment by parasidose.

Table (6): Show effect of parasidose on dogs infested with ticks and repellent effects of mixtures of essential oils on ticks with compared with control.

	% of attachment	% of dead ticks	% of dead ticks after retreatment (after 1 hour)	Repellent effect of ticks (reinfestations after (Days))
Pitbull	17.33± 5.38 ^a	82.65± 3.4 ^b	100± 0 ^a	2.95± 0.5 ^a
Rottweiler	47.5± 14.8 ^{ab}	86.0± 2.8 ^b	100± 0 ^a	2.6 ±0.58 ^a
Pitbull (control)	41.67±7.26 ^{ab}	0± 0 ^a	0± 0 ^b	0±0 ^b
Rottweiler (Control)	53.5± 12.07 ^b	0± 0 ^a	0± 0 ^b	0±0 ^b

Table (7): Show effect of parasidose on dogs infested with fleas and repellent effects of mixtures of essential oils on fleas with compared with control.

	% of attachment	% of dead fleas	% of dead fleas after retreatment after1(hour)	Repellent effect of fleas (reinfestations after (Days)
Pitbull	47± 2.38 ^a	94.46±0.22 ^b	100± 0 ^b	2.75 ±0.56 ^a
Rottweiler	54± 14.7 ^a	98.55± 0.84	100±0 ^b	2.577 ± 0.577 ^a
Pitbull (control)	56± 11.69 ^a	0± 0 ^a	0±0 ^a	0± 0 ^b
Rottweiler (Control)	55.5 ± 10.8 ^a	0± 0 ^a	0± 0 ^a	0±0 ^b

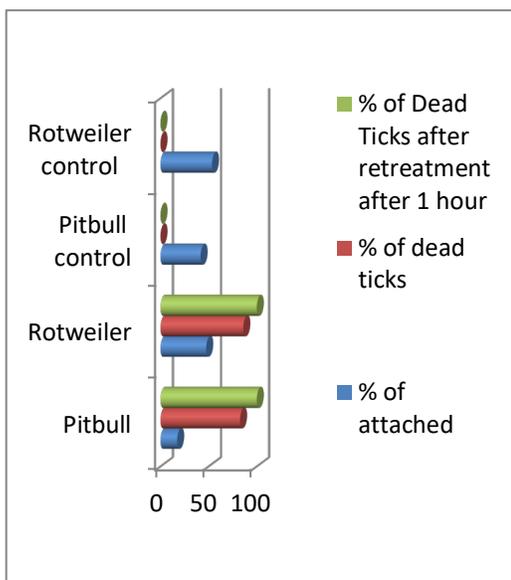


Fig. (6): Show effect of parasiticide on dogs infested with ticks and repellent effect of mixture of essential oils with compared with control.

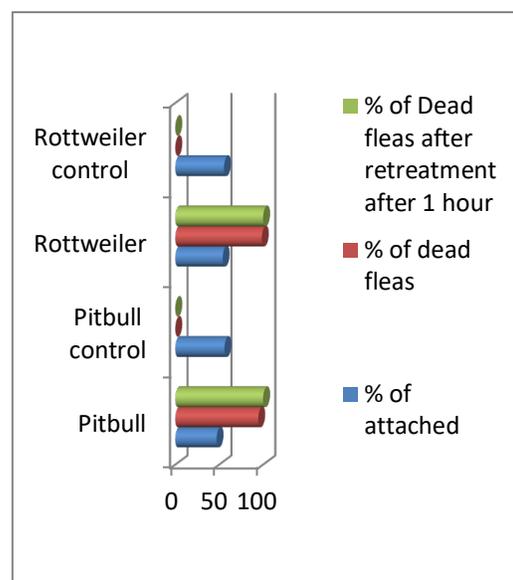


Fig. (7): Show effect of parasiticide on dogs infested with fleas and repellent effect of mixture of essential oils with compared with control.

DISCUSSION

In the present study, ectoparasites were collected from dogs in Egypt. The most common ones were ticks (*R. sanguineus*), Fleas (*Ctenocephalides canis* and *C. felis*). *C. felis* was reported to be the most abundant flea in dogs in Egypt, this result similar to **Tavassoli et al. (2010)**, Spain **Gracia et al. (2008)** in Iran. During examination of 117 dogs, % of tick infestations was the highest, these results were similar to those mentioned by **Heukelbach et al. (2012)**

who showed that among the ectoparasites, and (89.7 %) dogs were infested with *R. sanguineus*, (74.2 %) with *C. felis*. Among age of dogs, the highest infection rate with ticks and fleas was recording in dogs less than 1 year these results were similar to those mentioned by **Shoorijeh et al. (2008)**, **Xhaxhiu et al. (2009)**, **Chee et al. (2008)**, **Omudu et al. (2010)** and **Bensignor et al. (2014)**. Regarding to dog's sex, Percentage of flea and mixed infestations were higher in male than female, Results were similar to those reported by **Omudu et al. (2010)**, **Mosallanejad et al. (2011)**, **Jamshidi et al. (2012)** and **Hernández-Valdivia et al. (2011)** **Xhaxhiu et al. (2009)**. However, they added that there was no difference in the prevalence of infestation between sexes. Concerning breeds the highest prevalence level of those parasites was recorded in German shepherd dogs. Such result might be attributed to the increased popularity of this breed in Egypt than others. Moreover **Zabel and Hensel (2012)** added that, the high pH of the skin was followed by Rottweiler then American Pitbull. The obtained results were similar to those mentioned by **Bahrami et al. (2012)**, **Jamshidi et al. (2012)** and **Omudu et al. (2010)**. Regarding seasonal variation the highest percentage of ticks and fleas in dog was recorded in spring. Such result may be attributed to the increased temperature after winter months. (**Gonzalez et al. 2003**). These results were similar to those mentioned by **Millan et al. (2007)** and against **Cruz-Vazquez et al. (2001)**, **Dryden and Rust (1997)** and **Farkas et al. (2009)** and **Durden et al. (2005)**. The most common clinical signs recorded in dogs infested by ticks and fleas was emaciation, anaemia and pale mucous membrane, itching, alopecia. This result matched to those mentioned by **Ugochukwu and Nnadozie (1985)**, **Bahrami et al. (2012)**, **Figueredo et al. (2017)** and **Varloud and Hodgkins (2014)**. By microscopical examination of fecal sample of dogs infested by ectoparasites reveal presence of *Dipylidium caninum* egg nest and *Ascaris* adult and egg in some cases. Such results were similar to those mentioned by **Lam and Yu (2009)** and **Farag (2016)**. The collected ticks were identified as *R. sanguineus* by using the morphological keys provided by **Walker et al. (2000)**, **Nuttal and Warbuton (1911a)**. Fleas were identified as *C. felis*, this result was agreed with **Amin (1966)**, **Ihrke (2009a)**, **Jasmin (2011)**, **Day (2012)**, **Halliwell (2014)** and **Tavassoli et al. (2010)** in Iran, Spain **Gracia et al. (2008)**, United State of America **Harman et al. (1987)**, **Durden et al. (2005)** but disagree with **Wall et al. (1997)**, **González et al. (2004)** and **Aldemir (2007)**. Concerning using of Parasidose shampoo in vivo and in vitro, parasidose is a natural product, non-toxic, safe to human and has a physical action on insects (**Combescot-Lang et al., 2015**) who tested it on human louse and there was

no toxicity neither to the environment nor to human beings. That is why the authors would like to recommend its use for control of ticks and fleas as a natural control. However, there was a significant value between the incidence in dogs infested with ticks and fleas treated by parasidose shampoo and control group at $P < 0.05$ and those was the first study recorded from Egypt. Concerning using of mixture of natural oils as a repellent to avoid ticks and flea's bites as those mentioned by **Gunther Nentwig (2003)**, **Tawatsin et al. (2001)** and **Ellse and Wall (2014)**. It was proved that it has a repellent effect last for 2-3 days after treatment.

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