

## **Evaluation of clinical efficacy of Herbal product- *Yespray* on dogs as Ectoparasitocidal agents**

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### **Abstract:**

The efficacy of herbal ectoparasiticide *Yespray* was evaluated in-vivo and in-vitro against ectoparasites of 40 dogs. The formulated concentration was effective in controlling ectoparasites at 12-14 body spray, at twice/week application, and effective in killing fleas, ticks and mites at 15min, 72hr, and 8hr of exposure respectively(*in vitro*).

**Key words:** *Yespray*, ectoparasites, Dogs

### **Introduction:**

Ectoparasite infestation in dogs and human residential places is a serious problem. The ectoparasites( ticks, fleas) and mites not only inflict direct damage to dogs by causing severe itching, dermatitis and blood loss (Srivastava and Sinha, 2000), they are also responsible for transmission of Zoonotic protozoan, bacterial and viral diseases of man and animals (Chakraborti, 1997). A number of synthetic parasiticides have been tried for the ectoparasites with variable efficacy and toxicity (Link, 1965). The synthetic parasiticides residues are of serious environmental concern leading to pollution and health hazards to man and animals. Herbal drugs have been widely evaluated as an alternative method of parasite control, aiming to slow development of resistance and obtain low-cost biodegradable parasiticides (Chagas *et al.*, 2012). Botanical pesticides have different properties from plant to plant, possessing various effects against ectoparasites, and they also have antifungal and antibacterial properties. Hence efficacy of Herbal ectoparasiticide Product *Yespray* was assessed in-vivo and in-vitro against commonly occurring ectoparasites of dogs and results were recorded.

### **Materials and Method:**

A total of 40 dogs of different age group, sex, and breeds that are naturally infested with ectoparasites( fleas, ticks,) and mites, presented to Veterinary college Hospital, Hebbal, Bengaluru with following clinical manifestation viz, alopecia, and pruritus were selected for the study. At the formulated concentration of *Yespray* (consisting of Lemon grass oil-3%, Palmrosa-2%, Tulasi oil-0.5% and Eucalyptus oil-2%) was used for spraying on the body of the animals selected for study, and owners were advised not to bath/wash the pet for one/two days after spraying the *Yespray* at different intervals. The criteria for the evaluation of efficacy of the product included the visual examination for apparent reduction in number of ectoparasites on the body of the animals, and reduction in the symptoms of alopecia, scratching and general improvement in health of animals. All observations were recorded at different number and interval of application of the product.

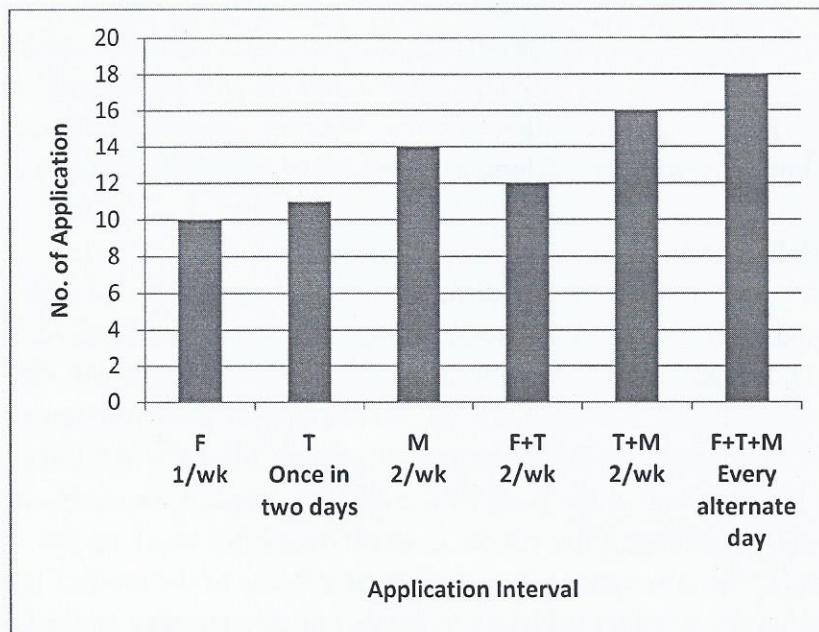
Acaricidal effect of *Yespray* against adult ticks was studied in vitro, and observations are recorded at 0hr, 24hr, 48hr, 72hr, and 96hr, after exposure for percent paralysis and mortality. Dog fleas collected were exposed by transferring them to vials which were freshly coated with *Yespray*, and percent paralysis and mortality were recorded at 0min, 5min, 10min, 15min, and 20min following exposure. In dogs infested with Mange/ mites, their skin scrapings were collected, and these were exposed to *Yespray*, later mites paralysis and mortality were recorded after 0hr, 2hr, 4hr, 6hr and 8hr of exposure under microscope.

Controls group with plain water exposure were kept for each batch/group of ectoparasites during study period.

**Results and Discussion:**

**Table 1: Profile for usage of Herbal spray (*Yespray*) on ectoparasites.**

Degree of infestation	Effective number Of applications	Effective application interval
Fleas(F)	10	Once a week
Ticks(T)	11	Once in two- days
Mites(M)	14	Twice a week
Fleas + Ticks(F+T)	12	Twice a week
Ticks + Mites(T+M)	16	Twice a week
Fleas + Ticks + Mites(F+T+M)	18	Every alternate Day

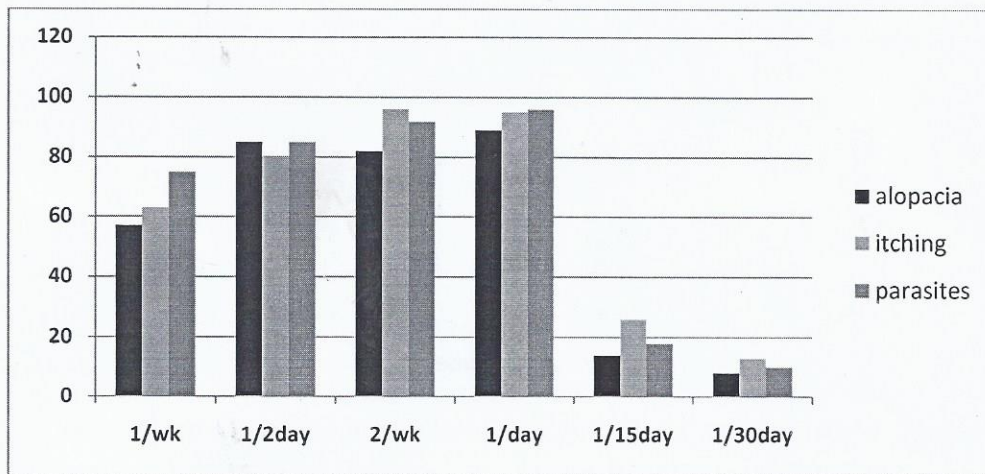


**Graph A: Profile for usage of *Yespray* on ectoparasites.**

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**Table 2: Effect of Yespray on alopecia, itching, degree of parasites,**

Application interval	Percent reduction		
	Alopecia	Itching	Parasites
1/week	57	63	75
1/2day	85	80	85
2/week	82	96	92
1/day	89	95	96
1/15day	14	26	18
1/30day	08	13	10



**Graph B: Reduction in alopecia, itching, and parasites at different application intervals.**

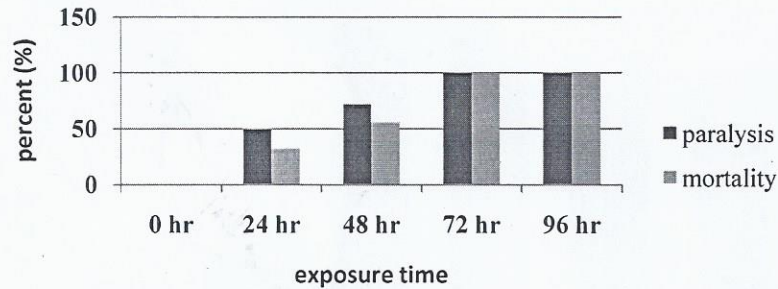
Age group	≤1yr	2-3yr	4-6yr	7-9yr	10-12yr	≥13yr
Effective number of applications	14	12	13	15	13	15
Effective application interval	2/week	2/week	1/2day	1/2day	1/2day	1/2day

Sex	male	Female
Effective number of applications	12	14
Effective application interval	2/week	2/week

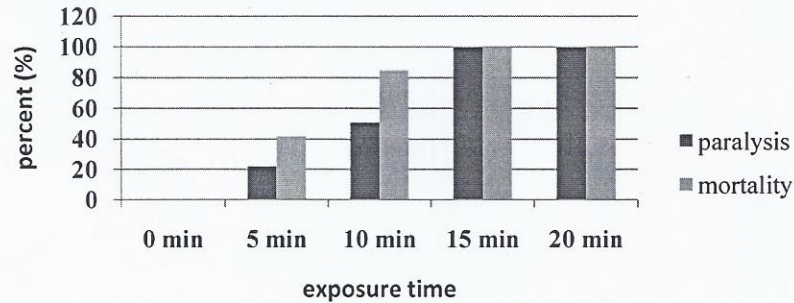
Breed of Dog	N/D	Lab	Pom	Pug	Gsd	C/B	C/sp	others
Effective number of applications	11	13	13	18	12	13	17	12
Effective application interval	2/week	2/week	1/2day	2/week	1/2day	2/week	2/week	2/week

**Table 3: In vitro effect of Yespray on Ectoparasites of dog.**

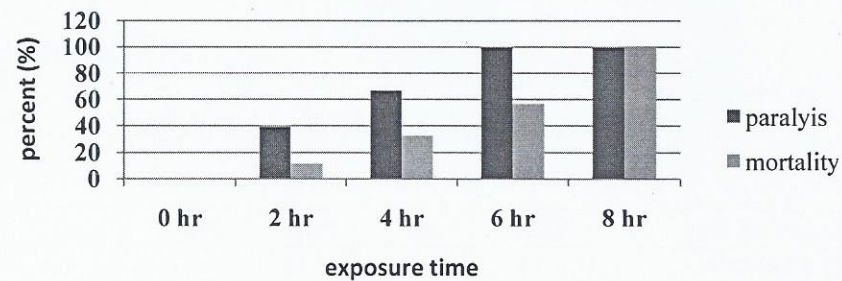
Ticks	Time (hr)	0	24	48	72	96
	Paralysis (%)	0	49	72	100	100
	Mortality (%)	0	32	56	100	100
Fleas	Time (min)	0	5	10	15	20
	Paralysis (%)	0	22	51	100	100
	Mortality (%)	0	42	85	100	100
Mites	Time (hr)	0	2	4	6	8
	Paralysis (%)	0	39	67	100	100
	Mortality (%)	0	12	33	57	100



**Graph 1 (a): In vitro tick paralysis & mortality at different time interval**



**Graph 2(a): In vitro fleas paralysis & mortality at different time interval.**



**Graph 3(a): In vitro mites paralysis & mortality at different time interval.**

The selected terpenes when mixed, with Citral (a component of certain essential oils, in particular lemongrass (*Cymbopogon citratus*), known to have limited antibacterial and anti-fungal effects) have been found to be highly effective, in aqueous solutions, in killing ticks and their eggs, fleas, mites and other parasites. The principal mode of action on adults and larvae is by direct solvent action on the wax-containing epicuticle of the chitinous exoskeleton, which is the primary means of controlling water loss in terrestrial arthropods.

At formulated concentration of *Yespray* as body spray, 12 to 14 number of applications at 2 spray per week (Table-1), gives effective control of both ticks and fleas. Kumar *et al.*, (2000) reported that Herbal ectoparasiticide AV/EPP/14, containing extracts of *Acorus calamus*, *Azadirachta indica*, *Pongamia glabra* (*Pongamia pinnata*), *Cedrus deodara* and *Eucalyptus globulus* sprayed five times at 6-day intervals (0, 6, 12, 18 and 24) resulted in elimination of 65.3, 87.6, 96.5, 99.6 and 100% of the ticks, respectively, and treated animals were free of ticks for a period of 30 days after the last treatment. Pathak *et al.*, (1998) found that the herbal ectoparasiticide AV/EPP/14, containing extracts of *Acorus calamus*, *Azadirachta indica*, *Pongamia glabra* (*Pongamia pinnata*), *Cedrus deodara* and *Eucalyptus globulus* at 1:4 concentration showed good acaricidal action, and the self-emulsifiable concentrate was easy to apply and non-irritant. All the dogs responded to treatment with no recurrence of infection in 20-25 days. These findings were consistent with our results that *Yespray*(2% Eucalyptus oil) has Acaricidal Property against ticks *in vivo*.

Das (2000) noted that a herbal preparation, which contains extracts of *Cedrus deodara*, *Azadirachta indica* and *Embelia ribes*, was applied as an aerosol spray to 14 stray and 5 pet dogs heavily infested with fleas (*Ctenocephalides canis*). A single application killed all of the fleas within 15 minutes and gave a slight improvement in the pruritic lesions, but had no effect on the allergic reactions to the flea bites and did not prevent rapid reinfection (shorter residual effect). These reports are in concurrence with our results that *Yespray* has biocidal effect on fleas both *in vivo* as well as *in vitro* studies.

On dogs, 10% combination showed 90% efficacy against *dog fleas* and nymphs of *R. sanguineus*, and 100% activity against larvae of *R. sanguineus*. Adults (especially males) of *R. sanguineus* were more tolerant to 10% combination; 3-4 applications were required to completely remove them (Srivastava, 1990).

Since mites are localized in skin layers and belongs to animals natural biota, with excessive proliferation rate that essentially parasitize hair follicle, sebaceous glands, its control requires more frequent applications at lesser duration of intervals.

Application of *Yespray* everyday, and twice a week has resulted in 89%, 95%, 96% and 82%, 96%, 92% reduction in alopecia, itching and number of parasites

respectively. (Table-2). With improvement in health condition of the treated animals, with gradual reduction in nasty odour of their environment. Daily and weekly treated animals showed clinical remission of lesions, possibly due to neem's anti-inflammatory and antibiotic properties (Fernandes, 2010) and eucalyptus oil as a natural anti-inflammatory, antiseptic action. Lemongrass oil (known to have limited antibacterial and anti-fungal effects) to impart fragrance there by making the mixture non-offensive.

Young age group dog tolerated application of *Yespray* well with no toxicity symptoms at given formulation of the product. With advancement of age and growth of animal more number of application are required with frequent intervals.

Thick haired breeds ( German Shepherd, golden retriever, Pomeranian, cocker spaniel) require frequent interval, more number of YESPRAY compared to less(coarse) haired breeds (N/D, Labrador, Pug), as thick hair may interfere with effective contact of product with parasite. No toxic side effects of *Yespray* were observed on treated animals at any of the doses.

After 24 hr exposure of *Yespray* (2% eucalyptus oil) to ticks in-vitro, (Table-3) 49% paralysis, with 32% mortality was seen. At 48hr and 72hr of exposure 72% paralysis, 56% mortality and 100% paralysis, and 100% mortality was noticed respectively, the results were concurrent with findings that in Eucalyptus there may be substances involved in a synergic Acaricide action that kills ticks in an average concentration of 17.5% (Chagas *et al.*, 2002).

At 5 min of exposure of *Yespray* to fleas (Table-3) 22% were paralysed, with 42% death. 100% paralysis and 100% mortality was observed at 15 min of exposure. The *Yespray* resulted into 39% paralysis, and 12% mortality of mites at 2hr of exposure, with 67% paralysis, 33% death at 4hr, and 100% paralysis at 6hr, 100% mortality at 8hr of exposure.

No paralysis and death were noticed in control group/batch of ectoparasites treated with plain water.

In order to reduce reliance on synthetic ectoparasiticides, there is need to diversify and evaluate eco-friendly compounds of plant origin. The efficacy of herbal formulations against mange in dogs (Das, 1996) and other ectoparasites in dogs has earlier been reported (Roy *et al.*, 1996).

Relatively longer exposure is required in comparison to synthetic chemicals (shorter residual effect). This may be viewed that plant based ectoparasiticides are not contact poisons but act through multiple modes of action, unlike synthetic ectoparasiticides that repel ectoparasites by the "hot foot effect" even after a very short contact, for only a few seconds, a "knock-down effect" occur and insects( ticks/fleas)

die soon after the open nerve ends at their feet got into contact with chemical (Elias, 2013). Though the action of Yespray in killing ectoparasites is somewhat delayed, owners opined that prolonged contact with the formulation is neither a discomfort nor a toxicity risk unlike chemical ectoparasiticides (Sharma, 1996).

Our study results were in concurrence with the findings of Robert *et al.*, (2000) that an antiparasitic formulation comprising in an antiparasitically effective amount of 5.3% *Eucalyptus oil*, 1.3% Cajeput oil, 2.0% *Lemongrass oil*, 2.5% Clove bud oil, 1.3% Peppermint oil, 2.7% piperonyl, 0.6% Piperonyl Butoxide, Surfactant 0.1% was effective in treating/preventing animal ectoparasitic infestation.

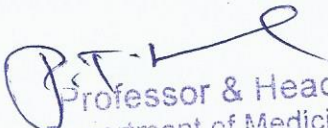
#### **Conclusion:**

In present study 40 dogs of different age, sex and breed were selected. Upon usage of formulated concentration of herbal ectoparasiticide Yespray as body spray, it was found to be effective in control of ticks, fleas, and mites at an average of 12 to 14 body spray, with twice-a-week application interval, with reduction in clinical Manifestations (alopecia, irritation and pruritus), and without any toxicity symptoms in younger group dogs. The results were supported by In-vitro study of Yespray, that formulated concentration of product was effective in killing fleas, ticks and mites at 15min, 72hr, and 8hr of exposure respectively. It is necessary to validate the in vivo use of safe and effective phytoparasitidal substances. Efforts should be focused on developing formulations that enhance the efficacy in vivo and lengthen the residual period.

#### **References:**

- Chagas, A.C.S., Barros, L. D., and Cotinguiba, F., 2012. In vitro efficacy of plant extracts and synthesized substances on *Rhipicephalus (Boophilus) Microplus* (Acari:Ixodidae) *Parasitology Research*, **110** (1): 295-303.
- Chagas, A. C. S., and Passos, W. M., 2002. Acaricide effect of *Eucalyptus* spp essential oils and concentrated emulsion on *Boophilus microplus* .*Braz. J. Vet. Res. Anim. Sci.* **39** (5).
- Chakraborti, A., 1997. A text book of preventive vety. medicine, 2<sup>nd</sup> Ed.:511-550.
- Das, S. S., 1996. Effect of herbal compound for the treatment of sorcoptic mange on dogs. *Vet.Parasitol.* **63**: 303-306.
- Das, S. S., 2000. Efficacy of pestoban aerosol spray against *Ctenocephalides canis* on dogs. *Indian Veterinary Journal* . **77** (4): 290-292.

- Elias, P., 2013. The use of Deltamethrin on Farm animals. Insecticides-“Development of Safer and more Effective Technologies” ed by Stanislav Trdan, ISBN978-953-51-0958-7.
- Fernandes J.I., Correia T.R.,and Ribeiro F.A., 2010. Efficacy of Neem (*Azadirachta indica*) in the control of *Demodex canis* on dogs *Revista Brasileira de Medicina Veterinária*, **32**(Supl.1):59- 63.
- Kumar R., Chauhan, P. P. S., Agarawal, R. D. and Daya Shankar., 2000. Efficacy of herbal ectoparasiticide AV/EPP/14 against lice and tick infestation on buffalo and cattle *Journal of Veterinary Parasitology* **14** No. (1): 67-69.
- Link, R. P., 1965. Insecticides on domestic animals in veterinary pharmacology and therapeutics edited by Jones L.M. Oxford and IBH publishing Co. Calcutta.
- Omar, S. F. A., and Sahar, E. R. S., 2014. Evaluation of Deltamethrin for control of some ectoparasites. *Egypt.J.Agri.Res.*, **92**(4):1596-3-1604.
- Pathak, K. M. L., and Shukla, R. C., 1998. Efficacy of AV/EPP/14 (herbal ectoparasiticide) against canine demodicosis. *Journal of Veterinary Parasitology*,**12**(1):50-51.
- Ryan, R. E., and Morris. S., 2000. Eucalyptus oil, cajeput oil, lemongrass oil, clove bud oil, peppermint oil, piperonyl and piperonyl butoxide; treating or preventing animal ectoparasitic infestation. US 6800294 B1.
- Roy, S., Maiti, S. K. and Ali, S.L., 1996. Therapeutic evaluation of herbal ectoparasites against canine demodicosis. *Indian.Vet. J.*, **73**: 871-873.
- Sharma, V., 1996. Efficacy of herbal formulation AV/EPP/14 against ectoparasites in pets. *Indian.Vet.Med.J.*,**20**: 11-16.
- Srivastava, P. S.and Sinha, S. R. P.1990. Ectoparasitocidal and acaricidal efficacy of the herbal insecticide - Pestoban - on cattle, buffaloes and dogs. *Pashudhan* . **5** (6): 4.
- Srivastava, P. S. and Sinha, S. R. P., 2000. Ectoparasites in livestock. *Livestock international*, **12**: 14-15.

  
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