

Comparative Studies on the Effect of Synthetic Pyrethroids and a Herbal Compound on Dogs Infested with Ticks



THESIS

SUBMITTED TO THE

RAJENDRA AGRICULTURAL UNIVERSITY

PUSA (SAMASTIPUR) BIHAR

(FACULTY OF POST-GRADUATE STUDIES)

In the partial fulfilment of the requirement

FOR THE DEGREE OF

Master of Veterinary Science

IN

(VETERINARY PARASITOLOGY)

By

Dr. Pankaj Kumar

Reg. No. - M/VPA/38/2002-2003

P. G. DEPARTMENT OF VETERINARY PARASITOLOGY

BIHAR VETERINARY COLLEGE

PATNA (BIHAR)

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DEDICATED
TO
MY DEAR PARENTS

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CERTIFICATE - I

This is to certify that Thesis entitled "**Comparative Studies on the effect of synthetic Pyrethroids and a herbal compound on dogs infested with ticks**" submitted in partial fulfillment of the requirements for the degree of Master of Veterinary Science (Veterinary Parasitology) of the faculty of Post-graduate studies, Rajendra Agricultural University, Pusa (samastipur) Bihar is the record of **bonafide research** carried out by *Dr. Pankaj Kumar* Reg. No. M/VPA/38/2002-2003 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

It is further certified that the assistance and help received during the course of this investigation and preparation of the thesis have been duly acknowledged.


(S. R. P. Sinha)
Major Advisor

Post-Graduate Department of Veterinary Parasitology
Bihar Veterinary College, Patna - 14
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CERTIFICATE - II

We, the undersigned members of the Advisory Committee of *Dr. Pankaj Kumar* Reg. No. M/VPA/38/2002-2003, a candidate for degree of Master of Veterinary Science with major in Veterinary Parasitology have gone through the manuscript of the thesis and agree that the thesis entitled "**Comparative Studies on the effect of synthetic Pyrethroids and a herbal compound on dogs infested with ticks**" may be submitted by *Dr. Pankaj Kumar* in partial fulfillment of the requirements for the degree.

(S. R. P. Sinha) 24/6/04

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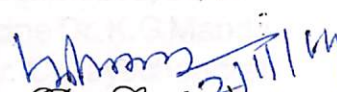
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CERTIFICATE - III

This is to certify that thesis entitled "**Comparative Studies on the effect of synthetic Pyrethroids and a herbal compound on dogs infested with ticks**" submitted by *Dr. Pankaj Kumar* Reg. No. MVPA/38/2002-2003, in partial fulfillment of the requirements for the degree of Master of Veterinary Science (Veterinary Parasitology) of the faculty of Post-graduate studies, Rajendra Agricultural University, Pusa (samastipur) Bihar was examined and approved on/...../2004.

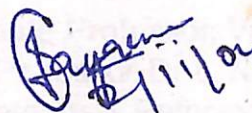

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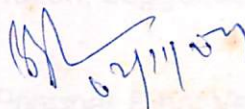
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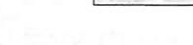
I am flooded with emotion and bow my head expressing profound sense of gratitude to my parents, brothers and sister for their constant encouragement, sacrifice and devotion without which it would not have been possible to carry out the present piece of work.

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Pankaj Kumar
24/6/04
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1

4



INTRODUCTION

Pet dogs are close companion to their owners, especially children and women acquire cozy nearness of their pets. External parasites such as ticks are very much prevalent among all the breeds of dogs. Ticks spend a part or whole of its life on the body of their host and so people owning or handling dog face high risk of exposure to the ticks and may acquire many zoonotic and tick transmitted protozoan, viral and bacterial diseases. Attachment of ticks also display unhygienic and clumsy appearance of dogs and ticks are suppose to one of the important constraint for the growth and development of its host. They not only create annoyance and worry to animals but also cause direct injury to the skin causing wounds which in turn lead to secondary infections. Their blood and lymph sucking habit during biting also inject certain toxins and antigens into the blood streams of their host. Therefore, it is necessary to control the ticks from the dogs as well as its surroundings such as cracks and crevices of kennel and underlighted, damp and swampy areas since ticks pass apart of their life cycle in the surroundings and the remaining part is completed over the host.

Despite many control and preventive measures, the control of ticks is mostly done by application of various acaricides. In order to choose the acaricides and its application certain difficulties pose great problems for control plans. Because ticks have high biotic potential and can survive under starvation for long periods. Their life cycle may include one, two or three hosts for feeding and reproductions. More over, the higher concentration of insecticide or acaricides cause toxic effects on dogs and the environment of dog lovers. The tick infesting dog species also develop and acquire high resistance to various acaricides due to their indiscriminate use. (Agrawal and Chauhan 2001). Intensive and continuous application of biodegradable

acaricides on dogs is coupled with hazardous effect to not only dogs but also human beings associated with immunosuppression.

There are four major groups of acaricides viz. chlorinated hydrocarbon, organophosphorus, carbamate and pyrethroids. Out of these, chlorinated hydrocarbons have very bad effect on environment and carbamate posses highly toxic properties with low margin of safety. Organophosphates are widely chosen but they get absorbed in the blood and have systemic effect on hosts. Pyrethroids, the new group, are extensively used for the control of ticks. Its member compounds deltamethrin and cypermethrin are well evaluated and reported to be highly effective against tick infestations. the choice between these two acaricides of the same group is still required to be evaluated because these are less toxic to the host and have minimum deleterious effect on the environment.

Apart from these chemicals, some herbal preparations of acaricides are also reported to posses high activity for the control of various ectoparasites including ticks. These herbal acaricides have qualities of easy application, safe use, almost no toxicity and high residual and no side effect to the environment. Despite of possessing very impressive and effective activity, there is a need to evaluate these preparations, particularly on dogs who remain in close contact with human beings specially to children. Since no comparative study has been undertaken to evaluate the herbal and pyrethroid compounds and also to compare within and between their groups of compounds on dogs infested with ticks.

With the advancement of modern technique of application and spraying of these acaricides, the most vulnerable problem is the unawareness about right dose and right use of these techniques. Many accidents occur as a result of improper use and application of overdose of acaricides. Similarly, sometimes underdosing found to

be inaccessible for required result.

Hence, it is urgent to study the degree of tick infestation in various breeds of dogs in local environmental condition for the control of various dogs ticks in urban Patna, which will be a guideline ^{for} effective control of ticks, not only betterment of dog fauna but also for the environment of pet lovers.

Most of the acaricides have been oil solvent and designed specially for spraying on walls and other inanimate objects and premises where the animal resides. The consequences of their application and indiscriminate use cause serious and toxic effects to the surrounding atmosphere and specially in case of pet dogs with confined indoor environment. The chances of insecticidal poisoning are greater in children and dogs, who is the victim of accidental ingestion if proper care should not be taken.

This is also required to study the adverse effect of higher doses and toxicity of acaricidal drugs to livestock including hematological and physiological parameters which may be utilised to acquire the knowledge of suitable therapeutic preventive measures, side effects and toxicity, diagnosis under the condition of accident. Keeping the above points in view, the present study was conducted with following objectives:-

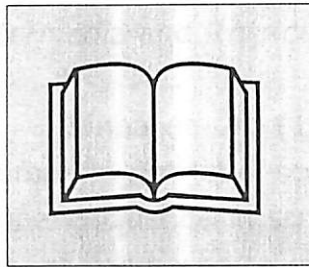
1. To identify degree tick infestation in various breed of dogs in and around Patna.
2. To evaluate the comparative efficacy of two pyrethroids, Deltamethrin and Cypermethrin and herbal compound on their recommended and slightly higher doses in dog infested with various species of ticks.
3. To find the toxic effects of treatments on dogs in respect to physiological and haematological parameters. ✍

REVIEW OF LITERATURE

Earlier report on dog use in India was by Spurr (1928) and Schöde (1948) after the survey done in 1938 and 1961 by Sar and brief resume of later studies on breed distribution of dog in India are given below.

(1) Studies on prevalence of dog

CHAPTER - 2 REVIEW OF LITERATURE



REVIEW OF LITERATURE

Earliest report on dog tick in India was reported by Sharif (1928) and Schulze (1935) after that detail survey done in 1938 and 1962 by Sen and Fletcher. A brief resume of later studies on prevalence and identification of dog ticks are as follows:-

(1) Studies on prevalence of dog ticks

Miranpuri and Singh (1978) noted prevalence of parasitic species of ticks were *Boophilus microplus* and *Rhipicephalus sanguineus* in cat , *Haemophysalis intermedia* and *Rhipicephalus sanguineus* in dogs.

Sinha *et. al.* (1987) found the tick of domestic animals in district of south and north Bihar and observed higher incidence in street dogs and lowest incidence in ass, as compared to cattle (73.61%) , goat (60.95%) , buffalo (59.75%) sheep (53.85%) and pet dogs (92.8%). The most common species of ticks prevalent in different district of Bihar were *Boophilus microplus*, *Hyalomma anatolicum*, *Hyalomma issaci*, *Rhipicephalus sanguineus* and *Haemophysalis bispinosa*. Seasonal incidence revealed the highest rate during the summer season (April - June) in all the animals while the lowest rate was during the winter (Nov. - Jan.).

Khan and Srivastav (1988) studied by model activity of *Rhipicephalus hemaphysaloides*, and noted that primary peak starts from middle of June and lasts upto November. While the secondary peak was of short duration (March to May). It infests chiefly dogs and buffaloes.

Mumcuoglu *et.al.* (1993) found a strong positive correlation between tick numbers and ambient temperature on dogs on parasitic stage of *Rhipicephalus sanguineus*. The mean percentage of dog infested in

winter month was 16.6 percent and increased in summer month to 34.4 percent. The male : female ratio was higher in winter (23:1) than in summer (1.1:1). The ear and abdomen were the summer predilection sites of ticks.

Inokuma *et. al.* (1995) studied the incidence of brown dogs tick in Okayama, Japan, in the summer dogs infested with *Rhipicephalus sanguineus*. showed pyoderma, anaemia, neutrophilia or Eosinophilia. The ticks were identified as *Rhipicephalus sanguineus*.

Dykstra *et. al.* (1997) surveyed veterinary clinics from urban and suburban areas of Texas for extent and source of tick related problems in dogs . In Texas cities prevalence of tick was between 15 and 21 per cent . The most frequently observed species was the brown dog tick, (*Rhipicephalus sanguine: us*). Tick infestation were commonest from May to August. Immature tick population were seen mainly in May and June and engorged female ticks in June and July.

Cruz *et. al.* (1999) studied seasonal distribution of *Rhipicephalus sanguineus* ticks on dogs in Morelos, Mexico and noted higher prevalence in spring, summer and autumn (20% or more) than in winter (13.7%). A positive correlation ($p < 0.01$) was found between prevalence of tick and rain fall plus temperature in spring, summer and autumn where as in winter there was only a correlation found with temperature.

Wilamowski *et. al.* (1999) reported that *Rhipicephalus sanguineus* was the most common ticks found in Israel town dogs. They further added several case reports mentioned attacks on man by *Rhipicephalus sanguineus* and other ticks.

Camacho *et. al.* (2003) studied various ticks from dogs in north west Spain by visiting clinics and indicated that *Ixodes hexagonas* clearly predominant over *Ixodes ricinus*.

Papazahariadou *et. al.* (2003) studied infestation of ticks in dogs in Thessaloniki (Greece) and found most prevalent species was (89.3%) *Rhipicephalus sanguineus* and (5.5%) *Rhipicephalus turanicus*. Out of them, 3.6% were nymphs and 1.6% were larvae and rest were adults. Dog living outdoors in rural areas and in close proximity to farm animals were infested with higher number of ticks than dogs living indoors. The most heavily infested sites were ear pinna, neck, interdigital skin folds. Cutaneous lesions at the attachment site were also noticed mainly in more severely infested animals.

2 . Studies on Hematological aspects on infestation of ticks and post-treatment

Goksu and Ozgencil (1970) found the lesions caused by adult *Rhipicephalus sanguineus* on the ear of sheep, which were hyperkeratosis with edematous swelling under the stratum germinativum, hemorrhages, and leucocytic infiltration around the blood vessels. Further observed hyperkeratosis was absent but a necrotic area at the attachment site surrounded by a region infiltrated with leucocytes made up of lymphocytes and red blood corpuscles (R.B.C) was there.

Tatchell and Moorhouse (1970) studied the biopsy sample of skin of dogs with attached ticks *Rhipicephalus* and observed the dilatation of blood vessels and hemorrhages in the region of tick attachment.

Springell and O'Kelly (1971) reported metabolic changes like reduction in total body water, decline in Hb, plasma albumin and plasma cholesterol level in tick infested steers were approximately the same as the amount of plasma and red cells consumed by ticks and concluded that the failure of tick infested animals to make up these losses, even after a very good diet, this may be due to the effect of tick toxins on the metabolic

process in tick infested cattle. However, the increase in plasma globulin contents was considered to be possibly due to the immunological response of the host.

Kelley *et. al.* (1971) reported reduction in the haematocrit haemoglobin in tick infested cattle and increase in the number of circulating lymphocytes and eosinophils and decrease in the neutrophil count.

Gabelhoff (1973) reported decrease in haemoglobin content and have adverse effect on erythropoiesis resulting an increase in mean corpuscular Haemoglobin concentration coupled with increased in the number of eosinophils and lymphocytes on either once or repeated infestation of nymphal / adult stages of ticks in calves, while the neutrophil were generally reduced in number of repeated infestation.

Ray (1980) showed an exaggerated pulse and respiratory frequency after pyrethroid treatment may be due to the release of catecholamine as result of action of Deltamethrin on adrenal medulla

McCorkie *et. al.*(1980) reported decrease in PCV, Hb, and TEC and increase in TLC in chicken, when they were treated with Deltamethrin spray in toxic doses.

Chinery (1981) detected of pharmacodynamic component of the tick saliva as a histamine blocking agent in the salivary gland of *Rhipicephalus sanguineus* which would provide an efficient mechanism for regulating the quantity of tissue fluid available for engorging tick. It also simultaneously neutralize the deleterious effect of excess pharmacodynamic substance produce in the host tissues.

Lawrence and Casida (1982) observed hyperthermic response (i.e. significant rise in rectal

temperature; femoral pulse and respiration rate) in pyrethroid treated rats and mice.

Maivisi *et. al.* (1986) measured biochemical constituents of sheep blood after giving Deltamethrin at 2 mg/ kg which caused inhibition of cholinesterase activity and unaffected concentration of Na, K and N.

Singh *et. al.* (1996) studied the haematology of skin biopsy samples were investigated in Dachshunds dogs with heavy (over 300 ticks of various stages), natural infestation of *Rhipicephalus sanguineus* compared with ticks free dogs. P.C.V., T.E.C., Hb levels were all significantly reduced in infested dogs. There were also significant change noted in the D.L.count. In infested dogs skin biopsy showed haemorrhagic spots, hyperanaemia, odema, inflammation and serous exudation at the attachment sites. The epidermis showed focal thickening with heavy infiltration of neutrophils and some lymphocytes.

Srivastava *et. al.* (2000) studied the effect of acaricides in different concentration on haemoglobin on different cross-bred calves infested with ticks and the study revealed that ivermectin at higher concentration significantly decrease the Haemoglobin as compared to control as well as recommended dose (0.2 mg/ kg body weight) at 24,48,72 hours of administration. Diazinon and deltamethrin significantly ($p<0.05$) decreased the Hb from control at all the three concentration (0.2, 3, 4 mg kg body wt.) within 24 hours of spraying.

Srivastava *et. al.* (2000) conducted a comparative studies on ivermectin, diazinon and deltamethrin application in different concentration in terms of total leucocyte count. The overall mean TLC value at various concentration of diazinon and deltamethrin showed significant ($p<0.05$) increase in average TLC value, however, there was no significant difference found among

mean values of different concentrations of ivermectin.

Szobo *et. al.* (2003) demonstrated that *Rhipicephalus sanguineus* considered to be natural parasite of dogs and is unable to develop appreciable resistance even after repeated feedings. Blood leucocytes and IgE levels in dogs were measured to detect a possible correlation with susceptibility or resistance of hosts. Since infested dogs develop an immediate hypersensitivity reaction to *Rhipicephalus sanguineus* antigen and total serum IgE also increased significantly after infestation. Infested dog did not display any alteration in blood count through out the experiment.

3. Evaluation of effect of acaricides (Synthetic Pyrethroids and Herbal Preparation against ectoparasites (Ticks)

Srivastava *et. al.* (1988) evaluated the ectoparasitidal properties of herbal preparation blaze (*A. calamus*, *C. deodara*, *E. certriodar* and *M. phillipinesis*) a shampoo form for the prevention and control of early infestation in dogs with ectoparasites. Preparation showed improvement in the symptoms of itching, biting of hairs, uneasiness and general appearance of body coat, at weekly intervals, total of 5 treatments extended over a month. Improvement in respect of quantum of reduction (81%) in larval and nymphal ticks were also seen in treated dogs.

Banerjee and Sangwan (1990) noted complete elimination of ticks on buffalos and cattle occurred following a single spray of butox (Decamethrin @25ppm) and these animals remain free from ticks at least 8-days. The drug had no ill effects on animals.

Khan & Srivastava (1990) tested that *Rhipicephalus sanguineus* was 1.322 and 1.2183 times more susceptible than *Rhipicephalus haemophysaloides*

to cyperkill and permasect respectively in an in-vitro trail. The inhibition and reproduction of treated females collates with the baseline data during peak activity period of *Rhipicephalus* ticks, even 0.05% wash of both the pyrethrin gave 93-98% control and protected treated dogs upto 11-15 days. With the proven ixodicidal competence these pyrethrins can be used either applying alternately with other compounds to avoid resistance or to monitor the resistant strain.

Srivastava and Sinha (1990) evaluated the efficacy of pestoban against ectoparasites those naturally infested the cattle, buffalo and dogs at Patna in Bihar. On dogs 10% pestoban showed 90% efficacy against nymphs of *Rhipicephalus sanguineus*, and 100% efficacy against larva of *Rhipicephalus sanguineus*. Adults (specially males) of *R. sanguineus* were more tolerant at 10% pestoban; 3-4 applications were required to remove them completely. No toxic side effects of pestoban were observed on treated animals.

Jani *et. al.* (1991) tried Butox (Deltamethrin) 0.0025% concentration against various stages of *Rhipicephalus sanguineus* in dogs of variable age, breed and sex. The cure rate was found to be 83.34% against ticks on single application repeat application on 7th day eliminated the ticks completely.

Tamang *et. al.* (1991) reported toxicity signs of Cypermethrin (a synthetic pyrethroid, by drenching with lethal dose, 600 mg/kg body weight) in goats such as grinding of teeth, waving of tail, dilatation of pupil and hyperaesthesia, excessive salivation, incoordination, dyspnoea, muscles, recumbency, coma and death (after 30 minutes).

Gatne *et. al.* (1992) : - treated 30 days with tick infestation (*Rhipicephalus sanguineus*) with amitraz (Tikoff - 12.5% E.C) at 180 ppm solution. The dogs

were German shaphered , Dobermann, Pomeranian, Boxer, Lhasa Apso and non-descript breeds. Two dogs had generalised and 28 had mild to moderate tick infestation. The drug was 100% effective in 26 cases, with only one application and two application in 4 cases. The drug was well tolerated by 27 dogs, but two Pomeranian and one Lhasa apso revealed side effects like milk seddation and pruritis which disappeared within 24 hours.

Khan and Srivastava (1992) evaluated synthetic pyrethroids cyperkill, permasect and sumicidin, both *in-vitro* and *in-vivo* against most prevalent ticks. Cyperkill was found most effective followed by permasect and sumicidin. Fertility of treated female (*in vitro*) upto the females replicated on treated animals upto 21 days was inhibited and resulted in sufficient destruction of subsequent generation. The period for protection from reinfested was 10 - 15 days with 0.05, 0.1 and 0.15% of these pyrethroids. The prolonged residual effect reduces the no. of treatment vis-a-vis cost of control operations.

Kumar *et. al.* (1992) studied the comparative efficacy of Ivermectin and pestoban against tick infested calves and reported that Ivermectin at the dose rate of 200 µg/kg body weight in S/C was 100% effective within 120 hrs while pestoban in the dilution rate of 1:5 was 95% effective within 96 hrs against *Boophilus* spp., *Hyalomma* spp. and *Rhipicephalus* spp.

Srivastava *et. al.* (1993) evaluated the deltamethrin (0.2%) *invitro* and *invivo* against the developmental stages of cattle tick. The 0.2% concentration of butox was found to be most effective in removing all the stages of tick species at 48 hrs. of post application. The drug was found least toxic and had a residual protection for 30-45 days post treatment . This was also found to be perfectly safe to the

environment and personal handling.

Maske and Bhilegaonkar (1995) tested ectozee spray against *Rhipicephalus haemaphysaloides*, *Linognathus vituli* and *Hippobosca maculata* (*H. variegata*) on cattle and Sarcopites on dogs. Ectozee 25% was found to be 100% effective within 12 hours, 24 hours, 8-min, 15-min, and 1 hour against nymphal and adult ticks, lice, fleas and mites respectively, while ectozee 100% spray was effective against nymphal ticks, adult ticks, lice, fleas and mites in 8 hrs, 12 hrs, 4-min, 5-min, and 30 min. respectively.

Kinjavelekar and Parai (1995) evaluated therapeutic efficacy of Butox in cross bred animals and on single application of drug at 2.5 ppm dilution, complete clinical and parasitological recovery within 7 days was noted any toxicities and longer residual effects were not observed.

Mishra *et. al.* (1996) described in their findings on acute toxicity of Deltamethrin (IR. 3R) - 3 (2,2 dibromovinyl) -2, -2 dimethyl cyclopropane caboxylate of (5) - alpha cyano-2-phenoxy benzyl) a potent photostable and biologically active synthetic pyrethroid. They reported on spraying of this insecticide at 0.05, 0.25 and 1 % concentrations to calves, increase in TLC, ALT, AST and Transaminase activity and decrease in PCV, Hb, TEC, Serum protein and albumin were noted. Further noted that buffalo calves sprayed with 1% Deltamethrin exhibited loss in body weight to the extent of 10 kg. but the animals regained their body weight within a month.

Tripathy *et. al.* (1995) used ectozee-50 against dogs infested with *Rhipicephalus sanguineus*, *Heterodoxus longitarsus*, (unspecified) fleas and *Hippobosca* spp; were sprayed daily (once in 24-hrs) till they were free from ectoparasites. In *in-vitro* trial the larvae, nymphs and adult ticks required a maximum 1

ay, 2 sprays and 4 sprays to kill the ectoparasites respectively. *Hippobosca* spp. died within 5-min. of spray. *In-vitro* study revealed 100% efficacy in removing larvae, nymphs and adult ticks by application of 2 sprays, 3 sprays and 4 sprays respectively at 24 hrs intervals. Re-infestation of ticks were observed in 30% of cases after 22 days.

Sharma (1996) tested 1:4 Formulation of AV/EPP/14 against ticks in dogs on 2 to 3 application with an interval of 7 to 10 days had the effort of knocking of 40-60% of ticks following body bath. When sample left to dry on body followed by combing much of remaining tick eliminated as they loosen their hold and anchorage on the skin. Preparations had no side effect on skin.

Singh (1997) tried the herbal ectoparasite AV/EPP/14 on dogs having ectoparasitic infestation including tick, mange, mites and *Demodex canis*. Almost (87%) dogs showed response and clinical control and majority (89%) required up to 3-applications at 5 days intervals.

Panda and Mishra (1997) conducted *in-vivo trial of herbal formulation AV/EPP/14* on dogs experimentally infected with nymphs and adults of the dog ticks *Rhipicephalus sanguineus* showed that single application with the drug at 1:2 dilution completely eliminated nymphs and reduced the adult population by 98% and 1:4 dilution killed 100% nymphs and 90% adults. In separate trial under field condition 93-95% and 87-90% efficacy was found on single application of 1:2 and 1:4 dilution respectively on all stages of canine Rhipicephalid ticks, the residual effect was 14-18 days and 10- 12 days respectively. The drug was well tolerated and treated dogs showed no adverse effects.

Bhilegaonkar and Maske (1998) Conducted a trial to evaluate the efficacy of herbal ectoparasites AV/EPP/14 *in-vitro* against ectoparasites in dogs. The formulation

at 1:4 concentration was effective in killing larva, nymph and adult ticks (*R. sanguineus*) in 5 hrs, 30 hrs, 72 hrs. respectively.

Talukdar *et. al.* (1998) applied deltamethrin (Butox) diazinon (Neocidol) as wash in goats infested naturally with *H. bispinosa*. Deltamethrin at 2.5 ppm concentration had 100, 84.66 and 72.86% effective against larvae, nymphs and adults respectively, while at 50 ppm concentration provided 100, 92.16 and 86.08% control of these stages. Residual effect at these two concentrations was seen upto 18-20 days and 25-30 days respectively. Diazinon at 0.06% concentration showed 53.43, 39.98 and 31.71% control of larvae, nymphs and adult respectively. Where as at 0.12% concentration the percentage of control were 100, 77.80 and 59.38 with residual effects of 7-8 and 15-20 days respective concentration of Diazinon treatment on dogs.

Estrada Pena and Ascher (1999) compared the efficacy of Fipronil with topical administration and amitraz with impregnated collar for prevention of experimental and natural infestation by brown dog tick and found that the both the acaricides inhibited attachment and feeding of ticks but amitraz had a significantly greater effect than fipronil on no. of larvae, feeding ticks, egg hatchability and larval viability, indicating partial ability to interrupt tick life cycle.

Kumar *et. al.* (2000) treated tick infested buffalo and cattle with herbal ectoparasiticide compound AV/EPP/14 (*Cedrus deodara* 0.2 g, *Pongamia glabra* 0.5 gm, *Azadirachta indica* extent 0.3 gm, *Eucalyptus glabrus* 0.2 gm and *Acarus calamus* 0.2 gm in emulsified base) at 1:4 was sprayed 5 times at 6 days interval (day 0, 6, 12, 18, 24) which resulted in elimination of 65.3, 87.6, 96.5, 99.6 and 100% of ticks respectively. The residual effect was found 30 days after last treatment.

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Agrawal and Chauhan (2001) given a detail

account of acaricides for control of ticks. According to the tick control plan it should be divided into two groups includes research applications while the other includes practical applications.

Srivastava *et. al.* (2001) conducted a trial on the efficacy of Deltamethrin (1.25% W/V), Cypermethrin (100 EC) & Fenvalerate (20% EC) in livestock animals against hard and soft ticks. Approximately 500 ml of each preparation at 2, 4 & 6 ml per litre in water sprayed uniformly. As per their observation, they recommended to avoid the higher dose application of Deltamethrin and Cypermethrin as they produce immense irritation and photosensitization symptoms at their higher doses.

They further suggested possible reason for the side effect, irritation & photosensitization may be the presence of cyclopropane ring as per their configuration of synthetic pyrethroid acaricides and pipernonyl butoxide (synergistic compound) enhances the neuralgic effect (e.g. photosensitization).

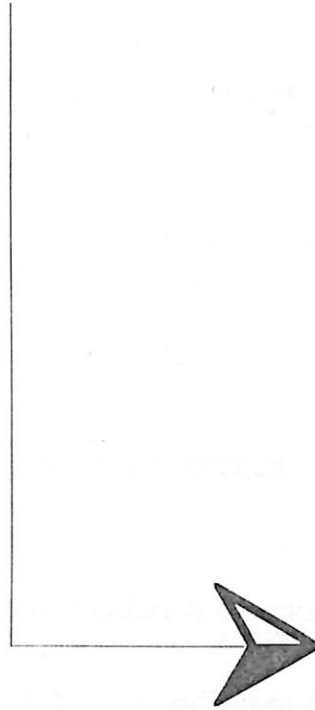
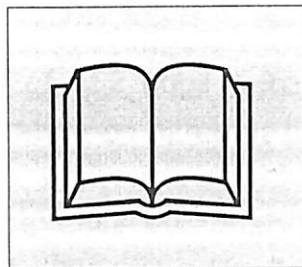
Endris *et. al.* (2002) evaluated the efficacy of 3 and 4 ml dose volume of tropically applied 65% permethrin against *Ctenocephalides felis* and *Rhipicephalus sanguineus* on dogs, their result indicated that a dose volume of 3 or 4 ml 65% permethrin is needed to obtain an adequate level and duration of efficacy against both *C. felies* and *R. sanguineus* on dogs weighing upto 30 kgs.

Vardharajan *et. al.* (2002) described that by relieving the animal from external parasite, the incidence of dreadful dieases transmitted through these parasites can be controlled. This saves the valuable animals from the onslaught of diseases and saves time and energy in treating the affected animal and basically external parasites are controlled by application of dipping, spraying, dusting, logging etc. but animal should protected from illeffect of insecticides.✍

MATERIALS AND METHOD

Research in common refers to a search for knowledge through systematic methods from collection of data, processing of data, presentation of data, interpretation of data, and the interpretation of data. The research process is a continuous process that involves the collection of data, processing of data, presentation of data, and the interpretation of data. The research process is a continuous process that involves the collection of data, processing of data, presentation of data, and the interpretation of data.

CHAPTER 3 MATERIALS AND METHODS



MATERIALS AND METHOD

Research in common refers to a search for knowledge through statistical methods right from collection of data, processing of data, presentation of data, analysis of data etc. to the interpretation of data. In other words, research refers to the systematic processes consisting of enumerating the problem, collecting facts and after systematic analysis, conclusions are drawn which are based on data.

(1) Study on incidence of tick in different breeds of dogs:

Total of 662 dogs were screened out to see the tick infestation on different breeds of dogs.

Source: Local veterinary hospital, private clinic, pet and street dogs within the Patna urban areas.

To Study:

- (1) Incidence of ticks in different breeds of dogs in Patna.
- (2) Effect of season on the incidence of ticks in dogs.
- (3) Age wise incidence of ticks in different breeds of dog.

(2) Collection and identification of tick species:

- (1) Ticks and their developmental stages i.e. larvae, nymph, adult were collected carefully with the help of ether soaked cotton swab and collected in glass tube and covered with thin muslin cloth. Some of the ticks also preserved in 70% alcohol.
- (2) Collected ticks were brought to the laboratory and

examine under dissecting microscope by restraining it on plasticin.

- (3) Permanent slides of different developmental stages of collected ticks were also prepared and seen under microscope for confirmation of identification of its species.

Preparation of slides :- Permanent mount of ticks were prepared as follows:-

- (a) Ticks preserved in 70% alcohol were passed serially through 50% alcohol, 30% alcohol and distilled water (5 to 10 minutes in each). The same procedure was adopted for the ticks preserved in formalin except that the specimens were initially washed thoroughly with tap water for about 6-8 hours.
- (b) The specimens were then transfer in 10% potassium hydroxide (KOH) for 48 hours to dissolve the internal organs.
- (c) The ticks were washed thrice in distilled water (30 minutes each) in order to remove excess KOH. The specimens were then slowly pressed with a fine pointed wooden splinter to remove the dissolved internal organs.
- (d) The specimens were then transferred through in each ascending grades of alcohol (30%, 50%, 70% and 90% alcohol) for a periods of 90 minutes.
- (e) The specimens were cleared in carbo-xylol (carbolic acid 1 part: xylol 3 parts) for 30 minutes, later passed through xylol (5 minutes) and finally mounted in Canada Balsam/ D.P.X. and then different species of ticks were identified on the basis of their morphological characters as described by Hoogstral

(1956) and Soulsby (1982).

(3) To evaluate the comparative efficacy of synthetic pyrethroid and herbal compounds on its recommended and slightly higher doses:

- (i) Moderate to highly infested dogs (40) with ticks were randomly selected for present experiment.
- (ii) The dogs were divided into 2- groups, each consisting 20 dogs for the trial of normal concentration and slightly higher concentration of acaricides.
- (iii) Again each group was divided into 4 subgroups, out of which three were for the trial of treatment of each acaricides and one was served as control.
- (iv) The samples of Cypermethrin, Deltamethrin and herbal compound were procured and doses were decided as per stated table below.

Table

Classifying Group	Fermacological Name/ Composition	Trade Name	Dose		Manufacturing Company	Price
			Reco- mmended	Slightly Higher		
Pyrethroid	Cypermethrin (1%)	Notix-CP	1ml/litre water (1ml=10mg)	1.25ml/lit water	Vet care division Tetragon chennie Pvt. Ltd. C7/22 KSSIDC I.E., Yelahanka Bangalore	28.00/ 15ml
Pyrethroid	Deltamethrin (1.25%)	Butox	2ml/litre water (1ml=12.5mg)	2.5ml/lit water	Intervet India Pvt. Ltd. G-16/1 MIDC, Tarapur Thane	22.00/ 15ml
Herbal	Cedrus deodara Pongami aglobra Azadirachia India Eucalyptus globulus Acarus calamus	Zerokeet	100ml/ 200ml water	125ml/ 200ml water	Dabur Ayurved Limited 22 Site IV Sahibabad, Ghaziabad	40.00/ 100ml

- (v) The no. of ticks (all stages larva, nymph, adult separately) on ears of each experimental dogs were counted before treatment and freshly prepared solutions (as per experimental plan) of each acaricides were applied on whole body of dog.
- (vi) The no. of ticks (all stages) were counted on 24 hours on post-treatment from previous observed area i.e. ears.
- (vii) To determine the residual effect the treated animal observed till they become reinfested with any stage of ticks.
- (viii) The % efficacy were determined on the rate of dropping off larvae, nymph and adult from the host on post-treatment, on the basis of following formula:-

$$= \frac{\text{No. of ticks (Pre-treatment)} - \text{No. of ticks (Post treatment)}}{\text{No. of ticks (Pre treatment)}} \times 100$$

(4) To study the effect on physiological and hematological parameters on use of recommended and slightly higher doses of acaricides in dogs:

- (1). The **temperature** (°F), **pulse** (per minute) and **respiration** (per minute) were recorded in each dog before treatment and at 6-hour, 12 hour, 24-hour and 48-hours of post treatment (previously randomly selected dogs in comparative efficacy of drug).

(2) Hematological studies:

Hematological studies were conducted (previously randomly selected dogs in comparative efficacy of drug)

immediately after collection of blood. For hematological examination, 2.5 ml of blood was collected from cephalic vein or recurrent tarsal vein of each dog in a clean and empty glass vial containing EDTA (Ethylene diamine tetra acetate) at the rate of 1mg/ml of blood as anticoagulant before treatment and changes were recorded after 24 hrs., 48 hrs., 72 hrs. and 96 hrs. of post treatment. However, PCV (%) and ESR (mm/hrs.) were recorded before treatment and only at 98-hrs. of post treatment. The whole blood was for the determination of:-

(i) Haemoglobin (Hb gram%/dl)

(ii) DLC (Differential leucocyte count)

- (a) Neutrophil (%)
- (b) Lymphocytes (%)
- (c) Eosinophil (%)
- (d) Monocytes (%)
- (e) Basophil (%) (Basophil count was found negative in all treatment group throughout the experiment).

For differential Leucocyte count (DLC), thin and uniform smear were prepared on clean and grease free slides. Smears were dried in air and properly stained and labelled.

(iii) TEC (Total erythrocyte count)

(iv) TLC (Total leucocyte count)

(v) Packed cell volume (P.C.V %)

(vi) Erythrocyte sedimentation rate (E.S.R.):-

Above studies were carried out as per the standard method described by Schalm *et. al.* (1975) and Sinha (1998)

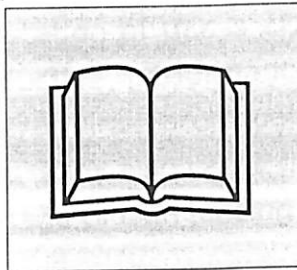
(v) Statistical Analysis:

The efficacy and toxicity of each acaricide on their normal and slightly higher dosage will be statistically analysed as per the method described by Snedecor and Cochran (1967).

RESULTS

Brown dog tick is being a great problem for pet owners and dog population. The current available information on the prevalence of brown dog tick in the pet population is very limited. The present study was conducted to determine the prevalence of brown dog tick in the pet population of the city of Addis Ababa, Ethiopia. The study was conducted in the city of Addis Ababa, Ethiopia, where the prevalence of brown dog tick was found to be 100%.

CHAPTER - 4 RESULTS AND DISCUSSION



RESULTS

Brown dog tick is being a great problem for pet lovers and dog population. The current available acaricides do not seem to give convincing results at the recommended dilution for eradication of ticks in dogs. Pyrethrin compounds like Deltamethrin and cypermethrin are most widely used in the Indian subcontinents but cannot be recommended at their higher doses as they produce immense irritation and photosensitization symptoms where as herbal acaricides are safe without any side effects but their operational performance supposed to be not up to mark. Infestation of tick is also of great concern as it affects production, performance of animals and transmission of several viral, protozoal and rickettsial disease to their host. They also cause blood loss, allergic responses and damage of skin leading to secondary bacterial infection.

So the present study was undertaken to see the incidence of various tick species infesting in different breeds of pet and stray dogs within the Patna urban areas. Further, comparative efficacy of cypermethrin, Deltamethrin and a herbal compound zerokeet were evaluated against natural tick infestation in dogs at their recommended and slightly higher doses. Moreover, post treatment responses on hematological and physiological parameters were also determined to evaluate the side effects and toxicity of these acaricides.

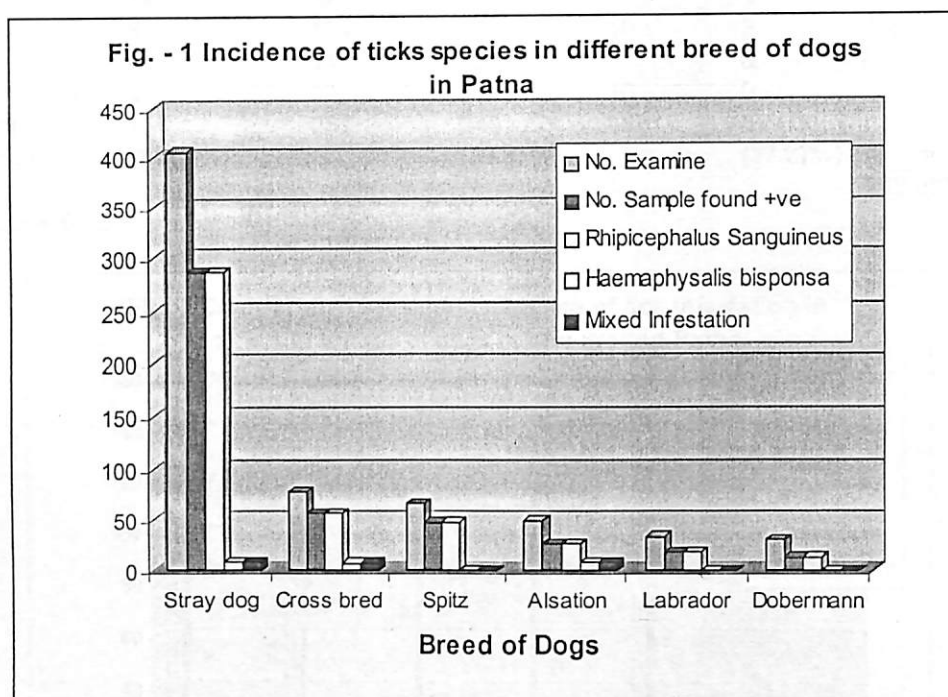
I. Incidence of Tick Infestation in different breeds of dogs

Altogether 662 dogs were screened in Patna and its surrounding areas to study the incidence of infestation of ticks. Out of which 257 and 405 were pet and stray dogs respectively. The overall positive samples were 445 (67.22%) (**Table 1**) and 70.37% of stray dogs were found positive with tick infestation. Maximum (71.79%)

infestation was found in cross-breed varieties where as 70.14%, 53.06%, 54.54%, 33.33% Spitz, Alsatian, Labrador, Doberman were infested with ticks respectively. All total positive dogs were infested with Brown dogs tick (*Rhipicephalus sanguineus*) where as infestation of *Hemophysalis bispinosa* was noted as mixed infestation (6.16%) with brown dog ticks only in stray, cross bred and Alsatian varieties of dogs.

Table -1 Incidence of ticks species in different breed of dogs in Patna

Breed of dogs	No. Examine	No. Sample found +ve	Tick species found		Mixed Infestation
			<i>Rhipicephalus sanguineus</i>	<i>Haemaphysalis bispinosa</i>	
Stray dog	405	285(70.37%)	285(70.37%)	8(2.8%)	8(2.6%)
Cross bred	78	56(71.79%)	56(71.79%)	7(12.5%)	7(12.5%)
Spitz	67	47(70.14%)	47(70.14%)	-	-
Alsatian	49	26(53.06%)	26(53.06%)	8(30.76%)	8(30.76%)
Labrador	33	18(54.54%)	18(54.54%)	-	-
Dobermann	30	13(33.33%)	13(33.33%)	-	-
Total	662	445(67.22%)	445(67.22%)	23(5.16%)	23(5.16%)



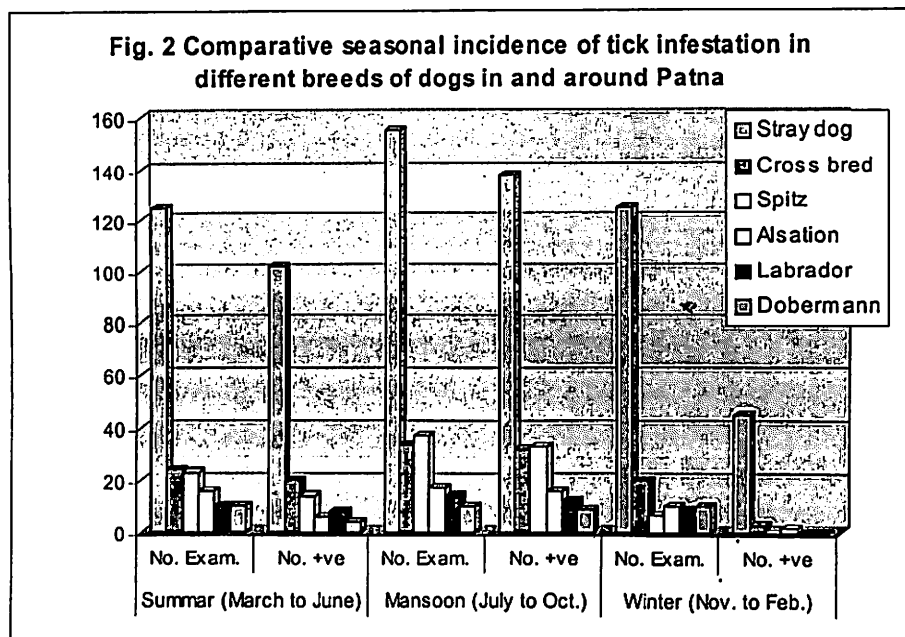
(a) Effect of season on incidence of tick infestation in and around Patna urban areas:-

Table 2, showing comparative seasonal incidence of tick infestation in different breeds of dog, which was found highest in monsoon (July - Oct.) 89.88%, followed by summer season (74.03%), where as, the least rate of infestation was noted in winter season (27.27%). This trend of seasonal incidence was same in all breeds of pet dogs as well as in stray dogs. The effect of season was found highly significant on the incidence of tick infestation in different breeds of dogs.

Table - 2 Comparative seasonal incidence of tick infestation in different breeds of dogs in and around Patna

Breeds of dogs	Summer (March to June)		Monsoon (July to Oct.)		Winter (Nov. to Feb.)		
	No. Exam.	No. +ve	No. Exam.	No. +ve	No. Exam.	No. +ve	
Stray dog	125	102	155	138	125	45	Total positive 445 (67.22%)
Cross bred	24	20	34	32	20	3	
Spitz	23	14	37	33	7	1	
Alsatian	16	8	17	16	10	2	
Labrador	10	6	14	12	9	0	
Dobermann	10	4	10	9	10	0	
Grand Total	208 (662)	154 (74.03%)	267	240 (89.88%)	187	51 (27.27%)	$X^2_{2df} = 202.08^{**}$

**** Significant at $p < 0.05$**



(b) Effect of age on the incidence of tick infestation in different breeds of dogs in and around Patna urban areas:-

The total 662 dogs examined were divided into 3 groups as per their age viz. 0-3 months, 3-12 months and more than 12 months (table-3) and the no. of sample examined 191, 213 and 258 respectively. Maximum (77.90%) incidence of ticks were noted in more than 12 months of age groups of all breeds including stray dogs. Slightly lower rate (73.70%) was observed between 3-12 months of age group and lowest (45.5%) infested animals were accounted under age group of 0-3 months or pups of all breeds of dog. The influence of age was found highly significant on incidence of tick infestation.

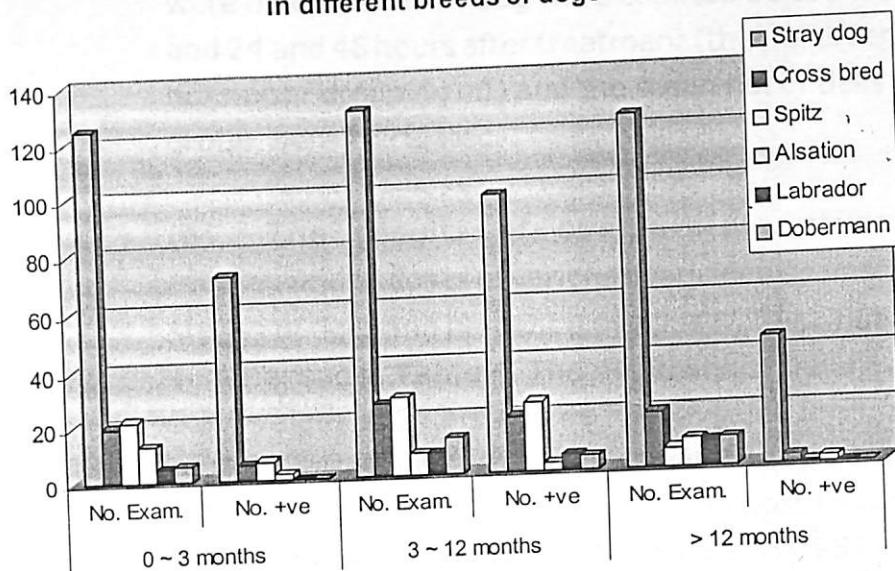
It was further noted that most favourable predilection sites for tick attachment were ears, eye, abdomen, back, scrotum udder and under the tail. During rigorous examination of their areas of predilection site, it was further noted that more male ticks dominated in these areas in respect of female ticks.

Table - 3 Comparative Age wise incidence of tick infestation in different breeds of dogs

Breeds of dogs	0 ~ 3 months		3 ~ 12 months		> 12 months	
	No. Exam.	No. +ve	No. Exam.	No. +ve	No. Exam.	No. +ve
Stray dog	125	72	130	98	150	117
Cross bred	20	6	26	20	32	28
Spitz	22	7	28	25	17	15
Alsatian	13	2	8	3	28	21
Labrador	5	0	8	6	20	12
Dobermann	6	0	13	5	11	8
Total(662)	191	87 (45.54%)	213	157(73.70%)	258	201(77.9%)
						$\chi^2_{2df} = 58.1^*$

* Significant at $p < 0.05$

Fig. 3 Comparative Study on Age wise incidence of tick infestation in different breeds of dogs



II. Comparative efficacy of Acaricides on its recommended and slightly higher doses against tick infested dogs:-

40 dogs between the age group of 10 months and above, naturally infested with ticks (*Rhipicephalus sanguineus*) were selected for the evaluation of comparative efficacy of two Pyrethroid compound Cypermethrin, Deltamethrin and herbal product Zerokeet at their recommended and slightly higher doses in two separate trials.

In first trial, the recommended doses were applied @ 1 ml/lit, 2 ml/lit and 100ml/200 ml; of Cypermethrin, Deltamethrin and Zerokeet respectively on randomly selected 3 groups of 5 dogs where one group (5 dogs) was kept untreated control throughout the period of observation.

Similarly in another trial slightly higher doses than recommended doses of same acaricides i.e. 1.25 ml/lit, 2.5 ml/lit, 125 ml/200 ml respectively on another three groups of same no. of dogs and one group was remained

untreated control throughout the period of experiment. The number of ticks (all stage larva , nymph and adult) were on ears of each dog were counted before treatment and 24 and 48 hours after treatment (the no. of remaining ticks after dropping off) and the mean no. of ticks of each group were calculated.

(a) The results of comparative efficacy on recommended doses of various acaricides against various stages of ticks and percent efficacies were evaluated and summarised in **Table 4**. The mean and standard error (\pm S.E.) values at 24 hrs and 48 hrs revealed that dropping of tick started just after application of each drug at its recommended dose. Within 24 hrs. on post treatment of cypermethrin, it was found 100%, 98.3% 95% against larval, nymphal and adult stages of ticks and within 48 hrs. cent percent ticks of all stages were dropped off. Similarly, Deltamethrin was found 75%, 89%, 87% effective against larva, nymph and adult stages of ticks respectively. Again in 48 hrs. nearly complete control of tick evaluated and the efficacy was found 99.5%, 99%, 98% respectively. Zerokeet was also able to 98%, 96%, 93% control of ticks larva, nymph and adult stages of ticks within 24 hrs of post treatment and 100% efficacies against each stages were noted within 48 hrs. of treatment.

Analysis of variance revealed that significant difference in decrease of larval stages ^{was found} within all treatment groups on 24 hrs post treatment. However, the differences were found to be non-significant in mean number of drop off nymph and adult stages within all treatment groups in 24 hrs. At 48 hrs of post treatment, significant decrease ($p < 0.05$) was again noted in Deltamethrin treatment group over control, whereas Cypermethrin and Zerokeet revealed 100 per cent efficacy.

The per cent efficacies also revealed the superior performance of Cypermethrin on its effectiveness and

Table -4 Comparative efficacy (%) of Acaricides on its recommended dose against tick infested dogs

Acaricides	No. of Dogs	Conc- entration	No. of ticks (Mean \pm S.E.) (Pretreatment)			No. of ticks (Mean \pm S.E.) (24 hrs Post treatment)			No. of ticks (Mean \pm S.E.) (48 hrs Post treatment)			Residual Effect (in days)
			Larva	Nymph	Adult	Larva	Nymph	Adult	Larva	Nymph	Adult	
Cypermethrin	5	1 ml/litre	42 ± 4.32	23.6 ± 2.24	12 ± 2.96	0.0 (100)	0.4 ^a ± 0.4 (98.3)	0.6 ^a ± 0.4 (95.0)	0.0 (100)	0.0 (100)	0.0 (100)	25-30 days
Deltamethrin	5	2 ml/litre	41 ± 4.63	21.6 ± 0.92	10.4 ± 1.74	10.6 ^a ± 0.6 (75.4)	2.4 ^a ± 1.85 (89.0)	1.4 ^a ± 0.4 (87.0)	0.2 ^a ± 0.2 (99.5)	0.2 ^a ± 0.2 (99.0)	0.2 ^a ± 0.2 (98.0)	15-20 days
Zerokeet	5	100 ml/200 ml	41.2 ± 5.89	20.4 ± 3.72	11.2 ± 1.15	0.8 ^b ± 0.37 (98.0)	0.4 ^a ± 0.4 (96.0)	0.8 ^a ± 0.37 (93.0)	0.0 (100)	0.0 (100)	0.0 (100)	20-30 days
Control	5		38.6 ± 4.29	18.6 ± 1.16	10.2 ± 1.52	38.6 ^c ± 4.29	18.6 ^b ± 1.16	10.2 ^b ± 1.52	38.3 ^b ± 4.39	18.9 ^b ± 1.18	10.2 ^b ± 4.39	

N.B. Mean with different superscripts (column wise) differ significantly at $p < 0.05$

quick control of all stages of ticks followed by Zerokeet which had found to be neck to neck effective with Cypermethrin but it has been slightly slower than previous one. Deltamethrin was also effective for the control of ticks at its recommended dose but values within 24 hrs of post treatment revealed slower and less effective performance than other two drugs.

To determine the residual effect, the treated animals were observed till they become reinfested with any stage of ticks. The mean residual protection period was noted highest (25-30 days) in Cypermethrin treated groups of dogs followed by Zerokeet treatment group (20-30 days) and than in Deltamethrin group (15-20 days).

(b) The result of comparative efficacy of slightly higher dose trial are summarised in Table no. 5.

The mean, \pm S.E. of number of larvae, nymph and adult ticks pre-treatment and after dropped off on applications of drugs are presented in **Table 5**. The result revealed 100 per cent efficacy of Cypermethrin just within 24 hrs. against all stages of ticks, similarly, Zerokeet also shown complete dropped off (100 per cent) of larvae and nymph stages, whereas 99% adults were controlled at its slight higher dose within 24 hrs. of treatment. Deltamethrin also found to be 98.6%, 95.23%, 90% efficacious against larva, lymph and adult stages. Within 48 hrs of treatment with slightly higher dose on the acaricides were found to be 100% effective for the control of all the stages of ticks. In this trial also superiority of Cypermethrin was noted to be highest followed by Zerokeet and Deltamethrin. Even residual performance of these Acaricides also confirmed the highest efficacy of the Cypermethrin (28-32 days), moderate of Zerokeet (25-30 days) and least of Deltamethrin (22-30 days).

Table -5 Comparative efficacy (%) of Acaricides on its slightly higher dose against tick infested dogs

Acaricides	No. of Dogs	Conc- entration	No. of ticks (Mean ± S.E.)			No. of ticks (Mean ± S.E.)			No. of ticks (Mean ± S.E.)			Residual Effect (in days)
			(Pretreatment)			(24 hrs Post treatment)			(48 hrs Post treatment)			
			Larva	Nymph	Adult	Larva	Nymph	Adult	Larva	Nymph	Adult	
Cypermethrin	5	1.25 ml/litre	38	23.6	14.2	0.0	0.0	0.0	0.0	0.0	0.0	28-32 days
			±5.41	±2.22	±2.87	(100)	(100)	(100)	(100)	(100)	(100)	
	5	2.5 ml/litre	43	21.4	10	0.6 ^a	1 ^a	1 ^a	0.0	0.0	0.0	22-30 days
			±4.80	±1.63	±0.94	±0.4	±0.31	±0.44	(100)	(100)	(100)	
						(98.6)	(95.23)	(90)				
Zerokeet	5	125 ml/200 ml	41.8	20.2	11.8	0.0	0.0	0.2 ^a	0.0	0.0	0.0	25-30 days
			±5.34	±2.23	±0.37	(100)	(100)	±0.2	(100)	(100)	(100)	
								(99)				
Control	5		40.4	21.7	10.2	40.4 ^b	21.7 ^b	10.2 ^b	40.4	21.7 ^b	10.2	
			±1.77	±1.91	±1.06	±1.77	±1.91	±1.06	±1.77	±1.91	±1.06	

N.B. Mean with different superscripts (column wise) differ significantly at p < 0.05

III. Study on physiological & haematological parameters after the use of different acaricides:-

The present study was carried out based upon previous trial on application of three acaricide at their recommended dose and slightly higher concentration. Physiological parameters viz. temperature, pulse, respiration rate were noted down pre treatment (0 hrs) and on Post treatment (6, 12, 24, 48 hrs) of each group of animals. The results on evaluation mean, S.E. and C.V.% of temperature, pulse and respiration rate at different time intervals are summarised in following tables and accordingly analysis of variance of each table were calculated to evaluate the significant changes within different treatments and time intervals.

(i) Effect of acaricides on temperature at different time intervals at recommended dose :-

The mean, S.E., and C.V.% of temperature at recommended concentration on spray of Cypermethrin, Deltamethrin and Zerkon on different time intervals are presented in **Table 6**. The overall mean temperature verses pooled over time intervals at recommended concentration show significant increase ($P < 0.05$) over control by 0.29 °F, 1.15 °F, 0.78 °F of Zerkon, Deltamethrin and Cypermethrin respectively. The maximum significant increase of temperature was noted on treatment with Deltamethrin followed by Cypermethrin and Zerkon treatments. The overall mean of time interval pooled over time interval, however revealed that changes were found to be non-significant at 6 hrs post treatment but over 0-hrs (pre treatment) but the temperature started increasingly significantly from 12 hrs and later successive periods of observations by 0.16, 0.43, 0.5, 0.5 of 6,12,24,48 hrs respectively.

Table - 6 Mean, S.E. and C.V. % of Temperature (°F) under different Acaricides (Recommended dose) at different time intervals in dogs

Name of Acaricides	Time Intervals		6-hrs		12-hrs		24-hrs		48-hrs		Overall Mean
	0-hrs Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	
Cyperm-ethrin	101.84 ±0.21	0.461	101.92 ±0.24	0.52	102.12 ±0.16	0.35	102.3 ±0.76	0.58	102.72 ±0.09	0.20	102.18 ^a
Deltam-ethrin	101.66 ±0.10	0.22	102.12 ±0.04	0.09	103.08 ±0.16	0.35	103.18 ±0.09	0.19	102.74 ±0.08	0.18	102.55 ^b
Zerokeet	101.64 ±0.231	0.50	101.64 ±0.22	0.48	101.68 ±0.32	0.71	101.76 ±0.16	0.35	101.76 ±0.36	0.80	101.69 ^c
Control	101.44 ±0.16	0.37	101.52 ±0.18	0.40	101.4 ±0.34	0.76	101.32 ±0.04	0.10	101.36 ±0.12	0.27	101.40 ^d
Overall Mean	101.64 ^a		101.8 ^a		102.07 ^b		102.14 ^{bc}		102.14 ^{abcd}		

Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column

Analysis of variance for the effect of these drugs at recommended dose concentration on body temperature at various time intervals have been depicted in **Table 7**. ANOVA revealed significant ($p < 0.05$) differences between time intervals and between treatments of drugs.

Table - 7 Analysis of variance of Temperature (°F) under different acaricide (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	6.19	34.98*
Between Time Interval	4	0.85	4.48*
Error	92	0.17	

* Significant at $p < 0.05$

(ii) Effect of acaricides on temperature at different time intervals at slightly higher dose:-

Similarly, mean, S.E. and C.V.% of temperature at slightly higher concentration on pre and post treatment of three experimental acaricides on dogs at different time intervals are presented in **Table 8**. The overall mean temperature verses pooled over time interval at slightly higher concentration revealed significant ($p < 0.05$) increase by 1.34 °F and 0.82 °F over control, on treatment with Deltamethrin and Cypermethrin respectively. However, changes between Zerokeet treated group and control group was found non significant but the overall temperature in Zerokeet group was lower than control group.

Table - 8 Mean, S.E. and C.V. % of Temperature (°F) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean			
	0-hrs Mean ±S.E.	C.V. %	6-hrs Mean ±S.E.	C.V. %	12-hrs Mean ±S.E.	C.V. %	24-hrs Mean ±S.E.	C.V. %
Cyperm-ethrin	101.9 ±0.17	0.37	102.62 ±0.06	0.13	103 ±0.2	0.42	103 ±0.06	0.13
Deltam-ethrin	101.64 ±0.11	0.24	102.76 ±0.23	0.50	103.54 ±0.09	0.21	103.88 ±0.32	0.69
Zerokeet	101.34 ±0.442	0.97	102 ±0.06	0.13	101.6 ±0.54	0.12	101.36 ±0.40	3.09
Control	101.76 ±0.11	0.24	101.74 ±0.10	0.23	101.7 ±0.17	0.39	101.72 ±0.12	0.26
Overall Mean	101.66 ^a		101.28 ^b		102.46 ^{bc}		102.44 ^{bcd}	

Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column

The trend of overall temperature changes at various time interval were slightly different as compared to changes observed with recommended doses, which also started increasing just 6 -hrs on post treatment with slightly higher dose but on later period of observation, the changes were found to be non-significant but trend started decreasing at 72 and 96 hrs on post treatment.

Analysis of variance for the effect of Cypermethrin, Deltamethrin and Zerokeet at their slightly higher concentration on body temperature at various time intervals have been depicted in **Table 9**. ANOVA revealed significant ($p < 0.05$) differences between time intervals and between treatment of drugs.

Table - 9 Analysis of variance of Temperature (°F) under different acaricide (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	14.87	63.82*
Between Time Interval	4	2.00	8.62*
Error	92	0.23	

* Significant at $p < 0.05$

(3) Effect of acaricides on pulse at different time intervals at recommended dose:-

The mean, S. E. and C.V.% for pulse/ minute at recommended concentration of drugs at different time intervals are presented in **Table 10**.

Table - 10 Mean, S.E. and C.V. % of Pulse (rate/min) under different Acaricides (Recommended dose) at different time intervals in dogs

Name of Acaricides	Time Intervals		6-hrs		12-hrs		24-hrs		48-hrs		Overall Mean
	0-hrs Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	
Cyperm -ethrin	81.6 ±1.16	3.18	85 ±1.13	2.98	88.6 ±1.12	2.82	92.8 ±0.85	2.06	88.8 ±0.37	0.93	87.36 ^a
Deltam- ethrin	83.4 ±0.92	2.48	90 ±0.31	0.78	94 ±1.14	2.71	95.6 ±1.36	3.18	93 ±0.94	2.27	91.2 ^b
Zerokeet	80.8 ±0.58	1.61	82 ±0.31	0.86	81.8 ±0.2	0.54	81.6 ±0.50	1.39	81.2 ±0.37	1.02	81.48 ^c
Control	81.6 ±0.50	1.40	81.8 ±0.37	1.02	81.6 ±0.4	1.09	81.6 ±0.4	1.09	81.4 ±0.24	0.67	81.6 ^{cd}
Overall Mean	81.85 ^a		84.7 ^b		86.5 ^c		87.8 ^{cd}		86.1 ^{be}		

Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column

The overall mean pulse rate pooled over time interval due to effect of recommended concentration of Zerokeet was found to be lower (non-significant) from control by 0.12 per min. Besides these, there was significant ($p < 0.05$) increase by 9.6 and 5.76 pulse/ minute were noted due to effect of Deltamethrin and Cypermethrin treatments respectively at their recommended dose.

However, the overall pulse rate started increasing significantly ($P < 0.05$) just within 6 hrs of post treatment and at 12 hrs again the change was noted to be significant ($P < 0.05$) over 0 hrs by 2.85 and 4.65/ min. However, at 24 hrs the increase was non-significant from observation of 12 hrs but at 48 hrs the pulse rate was started decreasing but changes were non-significant from previous post treatment observation.

The analysis of variance (**Table 11**) for pulse rate per minute has shown significant ($P < 0.05$) differences under various drug treatment at their recommended dose within treatment, time intervals and between time intervals.

Table - 11 Analysis of variance of Pulse (rate/min) under different acaricide (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	560.73	80.92*
Between Time Interval	4	105.21	15.19*
Error	92	6.92	

* Significant at $p < 0.05$

(4) Effect of acaricides on pulse rate at different time intervals at slightly higher dose:-

The mean, S.E. and C.V.% for pulse rate/min at slightly higher dose of acaricides at different ^{time} intervals are presented in **Table 12**. The overall mean pulse rate pooled over different treatment groups increased significantly ($p < 0.05$) by 8.06 and 5.04 on treatment Deltamethrin and Cypermethrin respectively. However, a decrease value by 0.12 /min was noted in Zerokeet group over control but change was noted to be non-significant between Zerokeet group and control group of animals.

The overall mean pulse rate on treatment with acaricides pooled over different time intervals increased significantly ($p < 0.05$) by 4.82/min at 6 hrs post treatment, again significant increase in the value of pulse were noted by 6.47, 8.5 on 12 and 24 hrs respectively of post treatment with slightly higher dose of drugs. At 48 hrs the overall pulse rate was found to be decreasing direction but it was non-significant from 6, 12 and 24 hrs of observations.

Analysis of variance of the effect of different drugs at slightly higher dose on pulse rate at various time intervals have been depicted in **Table 13**. Analysis of variance revealed significant ($p < 0.05$) differences between time intervals at various treatment, within time intervals and within treatments.

Table - 12 Mean, S.E. and C.V. % of Pulse (rate/min) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals								Overall Mean
	0-hrs Mean ±S.E.	C.V. %	6-hrs Mean ±S.E.	C.V. %	12-hrs Mean ±S.E.	C.V. %	24-hrs Mean ±S.E.	C.V. %	48-hrs Mean ±S.E.
Cyperm-ethrin	79.8 ±0.66	1.85	85.2 ±0.37	0.97	92.6 ±1.02	2.48	95.2 ±0.69	1.61	91.4 ±0.6
Deltam-ethrin	82.4 ±1.43	3.89	92.6 ±0.6	1.44	92.5 ±0.67	1.83	97.2 ±0.37	0.86	92.8 ±0.58
Zerokeet	82.0 ±0.31	0.86	84.4 ±0.4	1.05	85.0 ±0.54	1.44	83 ±0.83	2.25	82.2 ±0.58
Control	82.4 ±0.92	2.51	83.7 ±0.74	1.99	82.4 ±0.81	2.20	85.2 ±1.28	3.36	83.8 ±0.66
Overall Mean	81.65 ^a		86.47 ^b		88.12 ^c		90.15 ^{cd}		87.55 ^{bcd}

Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

Table - 13 Analysis of variance of Pulse (rate/min) under different acaricide (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	384.72	13.22*
Between Time Interval	4	204.34	7.02*
Error	92	29.08	

* Significant at $p < 0.05$

(5) Effect of acaricides on respiration rate at different time intervals at recommended dose:-

The mean, S.E. and C.V.% of respiration rate/minute at recommended concentration of drugs at different time intervals are presented in **Table 14**. The overall mean respiration rate pooled over time intervals on treatment of Deltamethrin and Cypermethrin increase significantly ($p < 0.05$) by 7.56 and 6.12 per minute from control respectively.

The changes were also noted to be significant within all three treatment groups. It was 1.44 between Deltamethrin and Cypermethrin and 8.04 between Zerokeet and Deltamethrin treatment groups. Again by 6.6 per minute difference were noted between Zerokeet and Cypermethrin treatment. However, the changes in Zerokeet group was found to be non-significant from control.

Table - 14 Mean, S.E. and C.V. % of Respiration (rate/min) under different Acaricides (Recommended dose) at different time intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean			
	0-hrs Mean ±S.E.	C.V.%	6-hrs Mean ±S.E.	C.V.%	12-hrs Mean ±S.E.	C.V.%	24-hrs Mean ±S.E.	C.V.%
Cyperm -ethrin	21.6 ±0.92	9.58	26.4 ±0.67	5.71	33 ±0.44	3.03	35 ±0.89	5.71
Deltam- ethrin	21.8 ±0.48	5.02	26.6 ±0.50	4.28	33.6 ±0.24	1.62	36.6 ±0.6	3.66
Zerokeet	21.2 ±0.37	3.94	23 ±0.31	3.07	23.2 ±0.2	1.92	22.8 ±0.37	3.66
Control	21.6 ±0.67	6.99	22.2 ±0.37	3.73	22.6 ±0.04	3.93	23.4 ±0.24	2.30
Overall Mean	21.55 ^a		24.55 ^b		28.1 ^c		29.45 ^d	

Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column

Respiration rate started increasing significantly ($p < 0.05$) from 6 hrs, 12 hrs, 24 hrs on post treatment by 3.0, 6.5, 7.9 per minute respectively, at 48 hrs decreasing value of overall respiration was noted from 24 hrs which was found to be significant ($P < 0.05$) but this decrease was non-significant with 12 hrs of post treatment however difference in values were significant ($P < 0.05$) at 48 hrs with 0 and 6 hrs of overall temperature.

The analysis of variance of the effect of different drugs at recommended dose on respiration rate at various time intervals have been depicted in **Table 15**. Analysis of variance revealed significant ($P < 0.05$) differences between time intervals at various treatment, within time intervals and within treatments.

Table - 15 Analysis of variance of Respiration (rate/mim) under different acaricide (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	427.32	126.15*
Between Time Interval	4	173.39	51.18*
Error	92	3.38	

* Significant at $p < 0.05$

(6) Effect of acaricides on respiration rate at different time intervals at slightly higher dose:-

The mean, S.E. and C.V.% of respiration rate per minute at slightly higher concentration of drugs at different time intervals are presented in **Table 16**.

Table - 16 Mean, S.E. and C.V. % of Respiration (rate/min) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals		6-hrs		12-hrs		24-hrs		48-hrs		Overall Mean
	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	
Cyperm-ethrin	22.6 \pm 0.50	5.04	34.8 \pm 1.01	6.55	37.2 \pm 0.69	4.13	35.6 \pm 0.74	4.69	32.8 \pm 0.48	3.32	32.6 ^a
Deltam-ethrin	21.8 \pm 0.37	3.83	31.8 \pm 0.48	3.44	37 \pm 0.54	3.30	38.4 \pm 0.50	2.96	36.8 \pm 0.2	1.21	33.16 ^a
Zerokeet	21.2 \pm 0.2	2.10	23.6 \pm 0.24	2.31	23.6 \pm 0.24	2.31	23 \pm 0.31	3.07	21.8 \pm 0.2	2.05	22.64 ^b
Control	21.4 \pm 0.50	5.32	22.6 \pm 0.24	2.42	22.0 \pm 0.31	3.21	22 \pm 0.31	3.21	22.4 \pm 0.24	2.44	22.08 ^{bc}
Overall Mean	21.75 ^a		28.2 ^b		29.95 ^{bc}		29.75 ^{bcd}		28.45 ^{bode}		

Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column

At 5% level of the respiration rate due to Zerokeet treatment didn't differ significantly over control while Deltamethrin and Cypermethrin showed significant ($P < 0.05$) increase over control by 11.08 and 10.52 respectively. The changes between Cypermethrin and Deltamethrin treatment group was however found non-significant. The maximum increase in the respiration rate was noted in Deltamethrin treatment group. At 5% level, the respiration rate increased significantly ($P < 0.05$) by 6.45 per minute at 6 hrs from control while successive increase noted in all periods of observations but the changes were found to be non-significant from 6 hrs of observations and at 48 hrs of treatment a decrease rate was noted from its previous observation.

The analysis of variance of the effect of different drugs at slightly higher dose on respiration rate at various time intervals have been depicted in **Table 17**. Analysis of variance revealed significant ($p < 0.05$) differences between time intervals at various treatments, within time intervals and within treatments.

Table - 17 Analysis of variance of Respiration (rate/mim) under different acaricide (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	924.86	105.04*
Between Time Interval	4	227.24	25.81*
Error	92	8.80	

* Significant at $p < 0.05$

HAEMATOLOGICAL STUDIES

Effects of Acaricides on Hb (gm%) on recommended dose of concentration:-

The mean, S.E., C.V.% of Hb (gm%) under recommended concentration at various time intervals are presented in **Table 18**. The overall mean Hb (gm%) pooled over time intervals due to effect of Cypermethrin and Deltamethrin decreased significantly ($p < 0.05$) over control by (0.97%) and (1.08%). While, non-significant difference was noted between the level of Zerokeet over control, however, a slight decrease was noted in Zerokeet group also (0.22%) from control group.

At recommended concentration Hb (gm%) decreased significantly ($p < 0.05$) at 24 hrs by 0.66% from 12 hrs. Again, in 48 hrs it increased non-significantly by 24 hrs of observation 0.03% however it was significantly decreased by 0.56% from 0 hrs of observations. Again, on 72 hrs and 96 hrs non-significant increasing trend was observed but (differences) were significant ($p < 0.05$) decrease from the 0 hrs observations.

The analysis of variance of the effect of different acaricides at their recommended concentration under various time intervals on Haemoglobin percent has been presented in **Table 19**. It reveals significant ($P < 0.05$) differences between time- intervals and action of drug individual and between treatment at their recommended dose concentration.

Table - 18 Mean, S.E. and C.V. % of Haemoglobin % (Gram %) under different Acaricides (Recommended dose) at different time intervals in dogs

Name of Acaricides	Time Intervals								Overall Mean
	0-hrs	6-hrs	12-hrs	24-hrs	48-hrs				
	Mean ±S.E.	Mean ±S.E.	Mean ±S.E.	Mean ±S.E.	Mean ±S.E.	C.V. %	C.V. %	C.V. %	
Cyperm-ethrin	11.52 ±0.26	5.11 10.24 ±0.25	5.54 10.32 ±0.26	5.78 10.38 ±0.25	5.58 10.48 ±0.22	4.73	10.58 ^a		
Deltamethrin	11.58 ±0.73	5.76 10.2 ±0.29	6.53 10.14 ±0.28	6.19 10.22 ±0.27	6.08 10.24 ±0.31	6.94	10.47 ^a		
Zerokeet	11.42 ±0.33	6.64 11.36 ±0.24	4.84 11.32 ±0.25	4.94 11.3 ±0.25	4.95 11.26 ±0.23	4.70	11.33 ^b		
Control	11.4 ±0.27	5.36 11.48 ±0.28	5.58 11.52 ±0.26	5.51 11.66 ±0.26	5.05 11.7 ±0.26	5.08	11.55 ^{bc}		
Overall Mean	11.48 ^a	10.82 ^b	10.825 ^{bc}	10.89 ^{bcd}	10.92 ^{bcd}				

Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column

Table - 19 Analysis of variance of Hb (Gram %) under different acaricide (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	7.15	39.72*
Between Time Interval	4	1.55	8.61*
Error	92	0.18	

* Significant at $p < 0.05$

Effects of acaricides on Hb (gm%) on slightly higher dose of concentration:-

The ~~overall~~ mean, S.E., C.V.% of Hb (gm%) at slightly higher dose of concentration than recommended dose of drugs at different time intervals are presented in **Table 20**. The ^{overall} mean Hb (gm%) pooled over time intervals due to effect of Cypermethrin and Deltamethrin were found to be decreased significantly ($p < 0.05$) over control by (1.54%) and (0.42%) respectively. However, in the Zerokeet group also decreased value by (0.08%) was noted over control group but the difference was found to be non-significant. Moreover, the changes in the Hb% on treatment with Zerokeet and Deltamethrin were also found to be statistically non-significant. Moreover, the changes in the values of Hb% on treatment with Zerokeet and Deltamethrin were also found to be statistically non-significant.

Table - 20 Mean, S.E. and C.V. % of Haemoglobin % (Gram %) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals								Overall Mean
	0-hrs Mean ±S.E.	C.V.%	24-hrs Mean ±S.E.	C.V.%	48-hrs Mean ±S.E.	C.V.%	72-hrs Mean ±S.E.	C.V.%	96-hrs Mean ±S.E.
Cyperm-ethrin	11.48 ±0.28	5.58	9.44 ±0.32	7.65	9.41 ±0.30	7.32	9.32 ±0.33	8.11	9.3 ±0.32
Deltamethrin	11.08 ±0.31	6.10	10.8 ±0.34	7.19	10.82 ±0.35	7.31	10.9 ±0.34	7.0	10.94 ±0.36
Zerokeet	11.46 ±0.35	6.98	11.36 ±0.24	4.84	11.28 ±0.27	5.49	11.14 ±0.23	4.75	11.04 ±0.23
Control	11.66 ±0.24	4.67	11.52 ±0.25	5.00	11.34 ±0.26	5.12	11.18 ±0.25	5.03	10.92 ±0.21
Overall Mean	11.42 ^a		10.78 ^b		10.71 ^{bc}		10.635 ^{acd}		10.55 ^{bcde}

Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column

Table - 21 Analysis of variance of Hb (Gram %) under different acaricide (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	10.03	17.91*
Between Time Interval	4	2.40	4.29*
Error	92	0.56	

* Significant at $p < 0.05$

At slightly higher concentration; Hb% (gm%) significantly decreased at 24, 48, 72 and 96 hours by 0.64, 0.71, 0.785 and 0.87% respectively over 0 hrs of observations. The value of Hb% between 24,48,72 and 96 hours were however, noted to be non-significant, at slightly higher dose concentration.

The analysis of variance of Hb (gm%) of the effect of various drugs at various time intervals as per experimental plan has been presented in **Table 21**. It revealed significant ($P < 0.05$) differences between time-intervals and drugs treatment at their slightly higher dose of concentrations.

Effect of Acaricides on TEC ($10^6/\text{mm}^3$) under recommended dose concentration:-

The overall mean, S.E. and C.V.% of TEC ($10^6/\text{mm}^3$) of drugs at different time intervals are presented in **Table 22**. The overall mean pooled over time interval due to effect of Cypermethrin and Deltamethrin significantly decreased over control group by 0.336 and $0.364 \times 10^6/\text{mm}^3$ respectively. These two values also found to

significantly ($p < 0.05$) lowered from the value of Zerokeet treatment group by 0.336 and $0.360 \times 10^6/\text{mm}^3$ respectively. In contrast, on post treatment of Zerokeet at its recommended dose concentration, TEC increases over control group non-significantly by 0.004.

At recommended concentration significant ($p < 0.05$) decrease in TEC was noted at 24 hrs and 48 hrs by 0.365 and $0.46 \times 10^6/\text{mm}^3$ over 0 hrs of observation. The values at 72 hrs however was also lower than 0 hrs but it had started increasing successively at 72 hrs and 96 hrs by $0.04 \times 10^6/\text{mm}^3$ and $0.145 \times 10^6/\text{mm}^3$ over 48 hrs of observation (non-significantly).

The analysis of variance of the effect of different drugs on TEC under different time intervals have been depicted in the **Table 23**. It revealed significant ($p < 0.05$) difference between time intervals, drugs and interactions between drugs at recommended dose concentration of acaricides.

Table - 22 Mean, S.E. and C.V. % of Total Erythrocyte Count ($10^6/\text{mm}^3$) under different Acaricides (Recommended dose) at different time intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean			
	0-hrs Mean ±S.E.	C.V. %	24-hrs Mean ±S.E.	C.V. %	48-hrs Mean ±S.E.	C.V. %	72-hrs Mean ±S.E.	C.V. %
Cyperm-ethrin	6.04 ±0.08	2.99	5.34 ±0.22	9.36	5.2 ±0.25	10.96	5.4 ±0.24	10
Deltam-ethrin	5.9 ±0.21	8.13	5.28 ±0.30	12.68	5.26 ±0.20	8.36	5.44 ±0.23	9.55
Zerokeet	6.06 ±0.08	2.97	5.96 ±0.07	2.85	5.78 ±0.17	6.74	5.6 ±0.35	14.28
Control	5.92 ±0.18	7.09	5.88 ±0.02	7.90	5.84 ±0.19	7.51	5.8 ±0.18	7
Overall Mean	5.98 ^a		5.615 ^b		5.52 ^{bc}		5.56 ^{bcd}	

Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

Table - 23 Analysis of variance of TEC ($10^6/\text{mm}^3$) under different acaricide (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	1.01	4.40*
Between Time Interval	4	0.66	2.90*
Error	92	0.23	

* Significant at $p < 0.05$

Effect of acaricides on Total erythrocytes count (T.E.C.)($10^6/\text{mm}^3$) on slightly higher dose of concentration:-

The mean, S.E., C.V.% of T.E.C. ($10^6/\text{mm}^3$) of drugs at different time intervals are presented in **Table 24**. The overall mean pooled over time intervals due to effects of Cypermethrin and Deltamethrin significantly decreased over control group by 0.448 and $0.52 \times 10^6/\text{mm}^3$ respectively.

These two values also found to be significantly ($P < 0.05$) lowered from the value of Zerokeet treatment group by 0.584 and $0.512 \times 10^6/\text{mm}^3$ respectively. In contrast, Zerokeet at its slightly higher dose concentration, TEC increases over control group non-significantly by 0.64.

Table - 24 Mean, S.E. and C.V. % of Total Erythrocyte Count ($10^6/\text{mm}^3$) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean			
	0-hrs Mean ±S.E.	C.V.%	24-hrs Mean ±S.E.	C.V.%	48-hrs Mean ±S.E.	C.V.%	72-hrs Mean ±S.E.	C.V.%
Cyperm-ethrin	5.98 ±0.08	3.01	4.6 ±0.25	12.39	4.98 ±0.23	10.24	5.18 ±0.28	12.16
Deltamethrin	5.7 ±0.18	7.33	4.68 ±0.33	16.02	5.0 ±0.21	9.4	5.06 ±0.32	14.22
Zerokeet	5.84 ±0.19	7.51	5.8 ±0.18	7.0	5.72 ±0.19	7.51	5.56 ±0.34	13.84
Control	5.78 ±0.17	6.74	5.6 ±0.35	14.28	5.72 ±0.19	7.51	5.6 ±0.35	14.33
Overall Mean	5.825 ^a		5.17 ^b		5.355 ^{bc}		5.35 ^{bed}	

Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

At slightly higher concentration, significant decrease in TEC was noted at 24 hrs by 0.655 over 0 hrs. and successive periods of observations but again at 96 hrs the value increased from previous 3 periods of observations but it was found non-significant. This change (96 hrs) was also found non-significant from 0 hr of the observation.

The analysis of variance¹⁰ of the effect of different drugs on TEC under different time intervals have been depicted in **Table 25**. It revealed significant ($p < 0.05$) differences between time intervals, drugs and interaction between drugs at slightly higher concentration of acaricides.

Table - 25 Analysis of variance of TEC ($10^6/\text{mm}^3$) under different acaricide (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	2.23	6.98*
Between Time Interval	4	1.18	3.71*
Error	92	0.32	

* Significant at $p < 0.05$

Effect of Acaricides on Total leucocyte count ($10^6/\text{mm}^3$) under recommended dose:-

The mean, S.E. and C.V.% of total leucocyte count at recommended concentration of drugs at different time intervals are presented in **Table 26**.

Table - 26 Mean, S.E. and C.V. % of Total Leucocyte Count ($10^3/\text{mm}^3$) under different Acaricides (Recommended dose) at different time intervals in dogs

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	
Cyperm-ethrin	12.9 \pm 0.36	6.20	14.6 \pm 0.25	4.45	14.84 \pm 0.29	4.38	14.64 \pm 0.27	4.09	14.2 \pm 0.13	2.04	14.236 ^a
Deltam-ethrin	12.94 \pm 0.36	6.33	14.46 \pm 0.30	4.63	14.64 \pm 0.27	4.09	14.2 \pm 0.13	2.04	13.88 \pm 0.24	3.96	14.024 ^a
Zerokeet	13.04 \pm 0.31	5.29	13.12 \pm 0.34	5.79	13.16 \pm 0.30	5.09	13.2 \pm 0.31	5.22	13.06 \pm 0.28	4.82	13.116 ^b
Control	12.94 \pm 0.10	1.77	13.12 \pm 0.08	1.44	12.94 \pm 0.36	6.33	13.04 \pm 0.31	5.29	13.14 \pm 0.22	3.72	13.036 ^{bc}
Overall Mean	12.955 ^a		13.825 ^b		13.895 ^{bc}		13.77 ^{cd}		13.57 ^{bcd}		

Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

The overall mean, TLC value pooled over time intervals on exposure of recommended concentration of Cypermethrin and Deltamethrin showed significant ($P < 0.05$) increase over control group by 1.2 and $0.988 \times 10^3/\text{mm}^3$ respectively. The value of Zerokeet, however also found to be increased over control group by $0.08 \times 10^3/\text{mm}^3$ but it was noted to be non-significant. While comparing the value of Zerokeet with cypermethrin and Deltamethrin it was found a significant decrease by 1.12 and $0.908 \times 10^3/\text{mm}^3$ respectively.

The TLC value due to effect of Cypermethrin, Deltamethrin and Zerokeet at recommended concentration at 24 hrs, 48 hrs and 72 hrs increased significantly ($p < 0.05$) over 0 hr by 0.87, 0.94 and $0.815 \times 10^3/\text{mm}^3$ respectively. However, at 96 hours a slight decrease value was noted but it was also significant ($p < 0.05$) over 0 hours by $0.615 \times 10^3/\text{mm}^3$. The TLC value at 24, 48, 72 hours, however, successively increased with advancement of time, but the values were non-significant between these intervals of time. Again at 96 hours, it has significantly slashed with its previous 72 hours observation but this decrease was also noted to be non-significant between all observed time intervals excluding 0 hours.

The analysis of variance of the effect of different drugs at its recommended concentration on TLC value at various time intervals has been depicted in **Table 27**. Analysis of variance revealed significant ($P < 0.05$) differences between time intervals and drugs and interaction between drugs and time intervals at recommended concentration of drugs.

Table - 27 Analysis of variance of Total Leucocyte Count ($10^3/\text{mm}^3$) under different acaricide (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	9.47	21.52*
Between Time Interval	4	2.91	6.62*
Error	92	0.44	

* Significant at $p < 0.05$

Effects of Acaricides on total leucocyte count ($10^3/\text{mm}^3$) under slightly higher dose concentration:-

The mean, S.E. and C.V.% of TLC at slightly higher concentration of drugs at different time intervals are presented in **Table 28**. The overall mean of TLC value pooled over time-interval on exposure of slightly higher dose concentration of Cypermethrin and Deltamethrin showed significant ($p < 0.05$) increase over control group by 1.976 and $1.808 \times 10^3/\text{mm}^3$ respectively. The value of Zerokeet, however also caused increase in the value over control group by $0.012 \times 10^3/\text{mm}^3$ but it was noted to be non-significant. While comparing the value of Zerokeet with Cypermethrin and Deltamethrin it was found a significant decrease by 1.796 and $1.964 \times 10^3/\text{mm}^3$ respectively.

Table - 28 Mean, S.E. and C.V. % of Total Leucocyte Count ($10^3/\text{mm}^3$) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	
Cyperm-ethrin	13 \pm 0.33	5.76	16.02 \pm 0.20	2.87	15.92 \pm 0.18	2.51	15.68 \pm 0.22	3.12	14.64 \pm 0.27	4.09	15.052 ^a
Deltamethrin	12.9 \pm 0.36	6.20	15.98 \pm 0.22	3.06	15.68 \pm 0.22	3.12	15.4 \pm 0.18	2.59	14.46 \pm 0.30	4.63	14.884 ^a
Zerokeet	12.94 \pm 0.36	6.33	13.04 \pm 0.31	5.29	13.2 \pm 0.31	5.22	13.14 \pm 0.22	3.72	13.12 \pm 0.34	5.79	13.088 ^b
Control	12.94 \pm 0.36	6.33	13.14 \pm 0.21	3.65	13.04 \pm 0.31	5.29	13.12 \pm 0.08	1.44	13.14 \pm 0.22	3.72	13.076 ^{bc}
Overall Mean	12.945 ^a		14.545 ^b		14.46 ^{bc}		14.335 ^{bcd}		13.84 ^{de}		

Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

The TLC value due to effect of Cypermethrin, Deltamethrin and Zerokeet at slightly higher dose concentration at 24 hrs, 48 hrs, 72 hrs and 96 hrs increased significantly ($p < 0.05$) over 0- hours by 1.6, 1.51, 1.39 and $0.895 \times 10^3/\text{mm}^3$ respectively. It was evident from the table that at slightly higher dose application of drugs, the TLC value increased maximum within 24 hrs and started decreasing successively (non-significant) from the 24 hrs onwards. It was found minimum at 96 hrs. But all these differences between time intervals from 24 hrs to 72 hrs were non-significant.

The analysis of variance of the effect of different drugs at its slightly higher concentration on TLC value at various time interval has been depicted in **Table 29**. Analysis of variance revealed significant ($p < 0.05$) differences between time-intervals and drugs and interaction between drugs and time-intervals at slightly higher concentration of drugs.

Table - 29 Analysis of variance of Total Leucocyte Count ($10^3/\text{mm}^3$) under different acaricide (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	29.76	47.23*
Between Time Interval	4	8.78	13.93*
Error	92	0.63	

* Significant at $p < 0.05$

**Effect of Acaricides on differential leucocyte count:
(All these values are calculated and analysed as per
the angle corresponding to mean % i.e. ARC SIN
value)**

(A) Lymphocyte (%) (On Recommended Dose)

The mean, corresponding values of Arc sin, S.E. and C.V.% of lymphocyte (%) at its recommended dose of drugs at different time-intervals are presented in **Table 30**. The overall mean lymphocyte (%) polled over time intervals due to effect of recommended dose of Zerokeet, Deltamethrin and Cypermethrin was increased significantly ($p < 0.05$) from control by 0.33, 0.72 and 1.03% respectively. The values are also found significant ($P < 0.05$) between Cypermethrin and Zerokeet group by 0.70% where as difference with Deltamethrin within other two group of treatment was found to be non-significant.

Lymphocyte percentage was observed increased non-significantly through out the period upto 72 hrs on post treatment. However, at 96 hrs the decrease in count was found significant ($p < 0.05$) from the control by 1.03%.

Analysis of variance (**Table 31**) revealed significant ($p < 0.05$) differences between time intervals and treatment of drugs at recommended dose on lymphocyte count.

Table - 30 Mean, S.E. and C.V. % of Lymphocytes (%) under different Acaricides (Recommended dose) at different time intervals in dogs

(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage)

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	
Cyperm-ethrin	27.80 \pm 0.72 (21.80)	5.79	27.81 \pm 0.69 (21.8)	5.53	28.64 \pm 0.57 (23.0)	4.46	29.72 \pm 0.45 (24.6)	3.43	30.78 \pm 0.38 (26.2)	1.75	28.95 ^a
Deltam-ethrin	28.38 \pm 0.27 (22.6)	2.11	28.51 \pm 0.25 (22.8)	1.99	28.38 \pm 0.34 (22.6)	2.71	28.78 \pm 0.45 (23.2)	3.47	29.19 \pm 0.38 (23.8)	2.98	28.64 ^{ab}
Zerokeet	28.09 \pm 0.45 (22.2)	3.63	28.23 \pm 0.55 (22.4)	4.42	28.51 \pm 0.39 (22.8)	3.08	28.23 \pm 0.55 (22.4)	4.42	28.23 \pm 0.55 (22.4)	4.42	28.25 ^b
Control	27.67 \pm 0.67 (21.6)	5.24	28.08 \pm 0.63 (22.2)	5.05	28.09 \pm 0.45 (22.2)	3.63	27.97 \pm 0.21 (22.0)	1.71	27.82 \pm 0.40 (21.8)	3.23	27.92 ^c
Overall Mean	27.98 ^a		28.15 ^{ab}		28.40 ^{ab}		28.67 ^{ab}		29.01 ^b		

N.B.-1. Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column
2. Figures in parenthesis indicate mean percentage of original values.

Table - 31 Analysis of variance of Lymphocyte (%) under different acaricide (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	5	3.87*
Between Time Interval	4	3.3	2.56*
Error	92	1.29	

* Significant at $p < 0.05$

(B) Lymphocyte (%) (On Slightly Higher Dose)

The mean corresponding arc sin value and C.V.% of lymphocyte% at slightly higher dose application of drugs at different time intervals are presented in **Table 32.**

The overall mean lymphocyte (%) pooled over time interval at slightly higher dose concentration due to effect of Cypermethrin increased significantly ($p < 0.05$) from control by 1.45% whereas a significant decrease was noted on treatment of Zerokeet from control by 0.56%. The group treated with Deltamethrin however, had a non-significant difference with control group but it was found significantly ($p < 0.05$) lowered by 0.99% from Cypermethrin group and higher by 1.02 % from Zerokeet.

Table - 32 Mean, S.E. and C.V. % of Lymphocytes (%) under different Acaricides (Slightly higher dose) at different time intervals in dogs

(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage)

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs	C.V. %	Mean	C.V. %	Mean	C.V. %	Mean	C.V. %	Mean	C.V. %	
	Mean		Mean		Mean		Mean		Mean		
	±S.E.		±S.E.		±S.E.		±S.E.		±S.E.		
Cyperm-ethrin	27.82 ±0.51 (21.8)	4.13	28.65 ±0.21 (23.0)	1.64	29.46 ±0.25 (24.2)	1.90	30.39 ±0.16 (25.6)	1.18	31.30 ±0.28 (27.0)	2.04	29.52 ^a
Deltamethrin	28.64 ±0.48 (23.0)	3.73	28.78 ±0.39 (23.2)	3.05	28.38 ±0.27 (22.6)	2.11	28.51 ±0.25 (22.8)	1.99	28.37 ±0.40 (22.6)	3.20	28.53 ^b
Zeroket	27.40 ±0.35 (21.2)	3.32	27.68 ±0.35 (21.6)	2.85	27.68 ±0.35 (21.6)	2.85	27.27 ±0.22 (21.9)	1.79	27.55 ±0.35 (21.4)	2.86	27.51 ^c
Control	28.09 ±0.45 (22.2)	3.63	27.96 ±0.43 (22.0)	3.46	28.10 ±0.39 (22.2)	3.16	28.10 ±0.13 (22.2)	1.06	28.10 ±0.13 (22.2)	1.06	28.07 ^{bd}
Overall Mean	27.98 ^a		28.26 ^a		28.40 ^a		28.56 ^a		28.83 ^a		

N.B. 1. Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

2. Figures in parenthesis indicate mean percentage of original values.

Table - 33 Analysis of variance of Lymphocyte (%) under different acaricides (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	18.08	21.27*
Between Time Interval	4	2	2.35 N.S.
Error	92	0.85	

* Significant at $p < 0.05$

It was observed that lymphocyte count increased with advancement of time but there was no significant changes noted within overall mean of lymphocyte count between different time intervals throughout the period of experiment. Analysis of variance (**Table 33**) of lymphocyte % under treatment of different acaricides at slightly higher dose at different time interval revealed a significant differences ($p < 0.05$) between treatment and was found non-significant between time intervals.

(C) Neutrophil (%) (Recommended Dose)

The mean, S.E., and C.V.% of neutrophil (%) at its recommended dose of drugs at different time-intervals are presented in **Table 34**. The overall Neutrophil (%) pooled over time-interval due to effect of Cypermethrin was found significantly decreased from control group by 1.34% significantly ($p < 0.05$). The changes between Deltamethrin and Zerokeet group were found to be non-significant from control group.

Table - 34 Mean, S.E. and C.V. % of Neutrophil (%) under different Acaricides (Recommended dose) at different time intervals in dogs

(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage)

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	
Cyperm-ethrin	53.98 \pm 0.84 (65.4)	3.48	53.37 \pm 0.61 (64.4)	2.56	53.13 \pm 0.68 (64.0)	2.86	52.54 \pm 0.68 (62.8)	2.89	51.35 \pm 0.49 (61.0)	2.12	52.87 ^a
Deltam-ethrin	54.33 \pm 0.42 (66.0)	1.74	54.21 \pm 0.22 (65.8)	0.92	54.21 \pm 0.35 (65.8)	1.43	54.09 \pm 0.36 (65.6)	1.47	53.73 \pm 0.32 (65.0)	1.35	54.11 ^b
Zerokeet	54.09 \pm 0.36 (65.6)	1.47	54.33 \pm 0.19 (66.0)	0.77	54.33 \pm 0.33 (66.0)	1.36	54.33 \pm 0.19 (66.0)	0.77	54.45 \pm 0.29 (66.2)	1.21	54.30 ^c
Control	54.46 \pm 0.52 (66.2)	2.14	54.57 \pm 0.40 (66.2)	1.66	54.45 \pm 0.48 (66.2)	1.96	54.09 \pm 0.41 (65.6)	1.68	53.49 \pm 0.23 (64.6)	0.99	54.21 ^{bcd}
Overall Mean	54.21 ^a		54.12 ^a		54.03 ^a		53.76 ^{ab}		53.25 ^b		

N.B. 1. Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column
2. Figures in parenthesis indicate mean percentage of original values.

The difference between Deltamethrin and Zerokeet was also found statistically non-significant but both treated group had significant ($p < 0.05$) higher value than cypermethrin group by 1.24% and 1.43% respectively.

At recommended dose significant ($p < 0.05$) decrease in neutrophil count was noted at 72 hrs and 96 hrs over previous three period of observation as Neutrophil % started decreasing from 24 hrs and throughout the period of experiment.

Table - 35 Analysis of variance of Neutrophil (%) under different acaricides (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	11.29	10.75*
Between Time Interval	4	2.73	2.6*
Error	92	1.05	

* Significant at $p < 0.05$

Analysis of variance of the effect of different drugs on Neutrophil count on application of recommended concentrations of drugs at various time-intervals have been depicted in **Table 35**. Analysis of variance revealed significant ($P < 0.05$) differences between five observation of time intervals and 3 drugs at their recommended concentrations.

Table - 36 Mean, S.E. and C.V. % of Neutrophil (%) under different Acaricides (Slightly higher dose) at different time intervals in dogs

(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage)

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	Mean \pm S.E.	C.V. %	
Cyperm-ethrin	53.86 \pm 0.77 (65.2)	3.23	53.85 \pm 0.44 (65.2)	1.81	52.77 \pm 0.14 (63.4)	0.60	51.70 \pm 0.04 (61.5)	0.19	51.0 \pm 0.47 (60.4)	2.07	52.63 ^a
Deltam-ethrin	53.62 \pm 0.79 (64.8)	3.30	53.50 \pm 0.84 (64.6)	3.51	54.21 \pm 0.40 (65.8)	1.64	54.09 \pm 0.49 (65.6)	2.01	54.70 \pm 0.49 (66.6)	2.01	54.02 ^b
Zerokeet	55.06 \pm 0.3 (67.2)	1.21	54.81 \pm 0.12 (66.8)	0.49	54.94 \pm 0.38 (67.0)	1.54	55.30 \pm 0.15 (67.6)	0.59	55.06 \pm 0.22 (67.2)	0.92	55.03 ^{bc}
Control	54.34 \pm 0.66 (66.0)	2.72	54.46 \pm 0.52 (66.2)	2.14	54.09 \pm 0.24 (65.6)	0.99	53.85 \pm 0.22 (65.2)	0.92	54.09 \pm 0.24 (65.6)	0.99	54.16 ^{cd}
Overall Mean	54.22 ^a		54.15 ^a		54.0 ^a		53.73 ^a		53.0 ^a		

N.B. 1. Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column
2. Figures in parenthesis indicate mean percentage of original values.

(D) Neutrophil (%) (At slightly higher dose)

The mean corresponding to Arc Sin values S.E. and C.V. % of Neutrophil (%) on post treatment of Cypermethrin, Deltamethrin and Zerokeet at their slightly higher dose concentration from recommended dose at different time intervals are depicted in **Table 36**. A significant ($p < 0.05$) decrease by 2.4% was noted in Neutrophil count on Cypermethrin treatment over Zerokeet group. Though decreased value also noted on treatment of Deltamethrin over control group by 0.14% but this was found non-significant change. Inverse effect observed on treatment with Zerokeet as Neutrophil count increased from control group by 0.87 but the difference was found to be non-significant. Neutrophil count were least in Cypermethrin treated group and highest on Zerokeet treatment.

The ANOVA Table for Neutrophil at slightly higher dose revealed effect of drugs at various time intervals has been depicted in **Table 37**. The table revealed non-significant ($p < 0.05$) difference between time-intervals and significant between drug interaction.

Table - 37 Analysis of variance of Neutrophil (%) under different acaricides (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	24.64	18.95*
Between Time Interval	4	1.09	0.84 N.S.
Error	92	1.30	

* Significant at $p < 0.05$

Table - 38 Mean, S.E. and C.V. % of Eosinophil (%) under different Acaricides (Recommended dose) at different time intervals in dogs

(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage))

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	
Cyperm-ethrin	18.58 ±0.70 (10.2)	8.44	18.42 ±0.42 (10.0)	5.15	18.40 ±0.60 (10.0)	7.33	18.24 ±0.19 (9.8)	2.35	18.03 ±0.38 (9.6)	9.3	18.33 ^a
Deltam-ethrin	18.03 ±0.38 (9.6)	4.71	18.03 ±0.38 (9.6)	4.71	18.41 ±0.51 (10.0)	6.19	18.03 ±0.38 (9.6)	4.71	18.24 ±0.19 (9.8)	2.35	18.14 ^a
Zerokeet	17.83 ±0.49 (9.4)	6.22	17.62 ±0.57 (9.2)	7.26	17.83 ±0.49 (9.4)	6.22	17.64 ±0.37 (9.2)	4.70	17.44 ±0.44 (9.0)	5.73	17.67 ^a
Control	18.20 ±0.63 (9.8)	7.80	17.83 ±0.49 (9.4)	6.22	17.82 ±0.59 (9.4)	7.40	18.24 ±0.19 (9.8)	2.3	19.17 ±0.34 (10.8)	3.96	18.25 ^a
Overall Mean	18.16 ^a		17.97 ^a		18.11 ^a		18.03 ^a		18.22 ^a		

N.B. 1. Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column
 2. Figures in parenthesis indicate mean percentage of original values.

(E) Eosinophil (%) (Recommended Dose)

The mean corresponding to Arc Sin, S.E. and C.V. % of Eosinophil (%) at recommended concentration at various time intervals have been presented in **Table 38**. The overall mean on Cypermethrin treatment was found 18.33% which was increased from control group (18.25%) by 0.08%. The value of Zerokeet and Deltamethrin decreased from the control by 0.58% and 0.11% respectively. Analysis of variance revealed interaction between all treatment groups were found to be non-significant. Similarly, differences from control group also found to be non-significant with all treatment group.

The analysis of variance (**Table 39**) of the effect of different drugs at their recommended concentration on Eosinophil count at various time intervals revealed non-significant differences between time-interval. It also revealed non-significant differences between treatment of drugs and interaction between treatments at various time-intervals.

Table - 39 Analysis of variance of Eosinophil (%) under different acaricides (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	2.2	2.17 N.S.
Between Time Interval	4	0.18	0.18 N.S.
Error	92	1.01	

* Significant at $p < 0.05$

Table - 40 Mean, S.E. and C.V. % of Eosinophil (%) under different Acaricides (Slightly higher dose) at different time intervals in dogs

(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage)

Name of Acaricides	Time Intervals								Overall Mean		
	0-hrs Mean ±S.E.	C.V.%	24-hrs Mean ±S.E.	C.V.%	48-hrs Mean ±S.E.	C.V.%	72-hrs Mean ±S.E.	C.V.%			
Cyperm-ethrin	18.78 ±0.63 (10.4)	7.50	17.85 ±0.24 (9.4)	2.96	17.85 ±0.24 (9.4)	2.96	18.03 ±0.38 (9.6)	4.71	17.85 ±0.24 (9.4)	2.96	18.07 ^a
Deltam-ethrin	18.58 ±0.70 (10.2)	8.44	18.37 ±0.81 (10.0)	9.85	18.42 ±0.42 (10.0)	5.15	18.60 ±0.46 (10.2)	5.59	18.04 ±0.23 (9.6)	2.93	18.40 ^a
Zerokeet	18.22 ±0.46 (9.8)	5.70	18.22 ±0.46 (9.8)	5.70	18.42 ±0.42 (10.0)	5.15	18.23 ±0.35 (9.8)	4.38	18.43 ±0.30 (10.0)	3.63	18.30 ^a
Control	18.23 ±0.35 (9.8)	4.38	18.22 ±0.46 (9.8)	5.70	18.24 ±0.19 (9.8)	2.35	18.24 ±0.19 (9.8)	2.35	18.24 ±0.19 (9.8)	2.35	18.23 ^a
Overall Mean	18.45 ^a		18.16 ^a		18.23 ^a		18.27 ^a		18.14 ^a		

N.B. 1. Overall mean with different superscripts differs significantly at p < 0.05 separately in row and column
2. Figures in parenthesis indicate mean percentage of original values.

(F) Eosinophil (%) (Slightly higher dose)

The mean, mean corresponding to arc sin, S.E. and C.V. % of eosinophil (%) at slightly higher concentration at different time intervals has been presented in **Table 40**. The values on treatment with Cypermethrin was found to be 18.07% which was decreased from control (18.23) by 0.16%. The value of Zerokeet and Deltamethrin increased from the control by 0.07 and 0.17 respectively. The analysis of variance revealed interaction between all treatment groups were found to be non-significant. Similarly the difference from control group were also found to be non-significant with all treatment groups.

The analysis of variance (**Table 41**) of then eosinophil values revealed drugs at their slightly higher concentration changes at various time intervals had non-significant differences. It also revealed non-significant differences in eosinophil count between treatment of drugs and interaction between treatment of drugs at various time intervals.

Table - 41 Analysis of variance of Eosinophil (%) under different acaricides (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	0.48	0.58 N.S.
Between Time Interval	4	0.30	0.36 N.S.
Error	92	0.82	

* Significant at $p < 0.05$

Table - 42 Mean, S.E. and C.V. % of Monocyte (%) under different Acaricides (Recommended dose) at different time intervals in dogs

(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage)

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	Mean \pm S.E.	C.V.%	
Cyperm-ethrin	8.87 \pm 0.45 (2.4)	11.38	8.5 \pm 0.36 (2.2)	9.6	8.5 \pm 0.36 (2.2)	9.6	8.022 \pm 0.67 (2.0)	18.70	8.392 \pm 0.78 (2.2)	20.73	8.45 ^a
Deltam-ethrin	7.652 \pm 0.47 (1.8)	13.85	7.174 \pm 0.58 (1.6)	18.13	7.174 \pm 0.58 (1.6)	18.13	7.174 \pm 0.58 (1.6)	18.13	6.696 \pm 0.58 (1.4)	19.43	7.17 ^b
Zerokeet	8.5 \pm 0.36 (2.2)	9.60	7.652 \pm 0.47 (1.8)	13.85	8.87 \pm 0.45 (2.4)	11.38	8.87 \pm 0.45 (2.4)	11.38	8.392 \pm 0.78 (2.2)	20.73	8.45 ^a
Control	8.392 \pm 0.78 (2.2)	20.73	8.022 \pm 0.67 (2.0)	18.70	8.022 \pm 0.67 (2.0)	18.70	8.704 \pm 0.97 (2.4)	25.05	9.24 \pm 0.45 (2.6)	10.93	8.47 ^a
Overall Mean	8.35 ^a		7.83 ^a		8.14 ^a		8.19 ^a		8.17 ^a		

N.B. 1. Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column
2. Figures in parenthesis indicate mean percentage of original values.



(4) Monocyte (%) (Recommended dose)

The mean, arcsin values, S.E. and C.V. % monocyte (%) at recommended concentration of drugs at different time-intervals was presented in **Table 42**. The overall mean monocyte (%) pooled over time-intervals due to effect of Deltamethrin found significantly ($p < 0.05$) decreased from control group by 1.3%. It (Deltamethrin) also significantly ($p < 0.05$) decreased from other two treatment groups by 1.28% (Cypermethrin and Zerokeet). However, the differences between Cypermethrin and Zerokeet group over control group (0.02) were found to be non-significant.

The analysis of variance (**Table 43**) revealed non-significant difference between time-intervals.

Table - 43 Analysis of variance of Monocyte (%) under different acaricides (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	10.39	6.14 *
Between Time Interval	4	0.71	0.42 N.S.
Error	92	1.69	

* Significant at $p < 0.05$

Table - 44 Mean, S.E. and C.V. % of Monocyte (%) under different Acaricides (Slightly higher dose) at different time intervals in dogs
(The values presented is in angle, corresponding to percentage (angle = Arc Sin Percentage)

Name of Acaricides	Time Intervals		24-hrs		48-hrs		72-hrs		96-hrs		Overall Mean
	0-hrs Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	
Cyperm-ethrin	8.87 ±0.45 (2.4)	11.38	8.87 ±0.45 (2.4)	11.38	9.24 ±0.45 (2.6)	10.93	9.61 ±0.36 (2.8)	8.53	9.24 ±0.45 (2.6)	10.93	9.16*
Deltam-ethrin	7.652 ±0.47 (1.8)	13.85	8.022 ±0.67 (2.0)	18.70	6.696 ±0.58 (1.4)	19.43	6.696 ±0.58 (1.4)	19.43	6.218 ±0.47 (1.2)	17.06	7.17*
Zerokeet	7.174 ±0.58 (1.6)	18.13	7.066 ±0.86 (1.6)	27.33	6.696 ±0.58 (1.4)	19.43	7.174 ±0.58 (1.6)	27.33	6.696 ±0.58 (1.4)	19.43	6.95*
Control	8.022 ±0.67 (2.0)	18.70	8.5 ±0.36 (2.2)	9.6	8.5 ±0.36 (2.2)	9.6	8.87 ±0.45 (2.4)	11.38	8.87 ±0.45 (2.4)	11.38	8.55*
Overall Mean	7.92*		8.11*		7.78*		8.08*		7.75*		

N.B. 1. Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column
2. Figures in parenthesis indicate mean percentage of original values.

(H) Monocyte (Slightly higher dose)

The mean, mean corresponding to arc sin values S.E. and C. V. % of Monocyte % at slightly higher concentration at different time-intervals is presented in **Table 44**. The overall mean Monocyte (%) pooled over time-intervals due to effect of Cypermethrin group increased significantly ($p < 0.05$) from control by 0.61%. The value of Monocyte (%) on treatment with Deltamethrin and Zerokeet group decreased significantly ($p < 0.05$) by 1.38% and 1.6% respectively over control group. The difference within Cypermethrin and Deltamethrin was also found to be significant ($p < 0.05$) by 1.99%. Again, on treatment of Zerokeet the values were noted 2.21 lowered from Cypermethrin treatment group, even difference in Zerokeet group was also found to be significantly ($p < 0.05$) lowered by 0.22% than Deltamethrin treated values.

The analysis of variance of different drugs at slightly higher concentration on monocyte count at various time-intervals have been depicted in **Table 45**. It reveals non-significant differences between time-intervals and significant differences in Monocyte count between drugs action and interaction between drugs at various time-intervals.

Table - 45 Analysis of variance of Monocyte (%) under different acaricides (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	30.13	21.22 *
Between Time Interval	4	0.55	0.38 N.S.
Error	92	1.42	

* Significant at $p < 0.05$

Effect of Acaricides on Erythrocyte Sedimentation Rate (E.S.R.) under recommended dose concentration

The mean, S.E. and C.V. % E.S.R. (mm/hrs) at recommended dose of drugs at 0-hours and 98-hours are presented in **Table 46**. The overall mean of E.S.R. (mm/hrs) values pooled over time-intervals on exposure to recommended concentration of drugs reflected increasing trend in all treatment group over control group but all these values were increased non-significantly by 0.06, 0.27 and 0.34 (mm/hrs) in Zerokeet, Deltamethrin and Cypermethrin treatment group respectively.

Table - 46 Mean, S.E. and C.V. % of E.S.R. (m.m./hour) under different Acaricides (Recommended dose) at different time intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean
	<u>0-hrs</u> Mean ±S.E.	C.V.%	<u>98-hrs</u> Mean ±S.E.	C.V.%	
Cypermethrin	5.9 ±0.19	7.28	5.96 ±0.14	5.53	5.93 ^a
Deltamethrin	5.84 ±0.21	8.04	5.88 ±0.22	8.33	5.86 ^a
Zerokeet	5.68 ±0.20	8.09	5.62 ±0.17	6.93	5.65 ^a
Control	5.42 ±0.29	11.99	5.76 ±0.23	8.85	5.59 ^a
Overall Mean	5.71 ^a		5.805 ^a		

N.B. : Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

Due to recommended dose of different drugs at 98-hours the E.S.R. value again found to be increased by 0.095 mm/hrs over 0-hour. But this increase was also found to be non-significant.

The analysis of variance of the effect of different

drugs at recommended concentration on E.S.R. values at 0-hours to 98-hours interval have been depicted in **Table 47**. The ANOVA revealed non-significant difference between drugs.

Table - 47 Analysis of variance of E.S.R. (m.m./hr) under different acaricides (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	0.26	1.26 N.S.
Between Time Interval	1	0.09	0.46 N.S.
Error	35	0.21	

N.B. = N.S. Non-Significant

Effects of Acaricides on Erythrocyte Sedimentation Rate under slightly higher dose concentration:-

The mean, mean corresponding to arc sin, S.E., and C.V. % of E.S.R. (mm/hrs) at slightly higher dose concentration of drugs at 0-hrs and 98-hrs are presented in **Table 48**. The overall mean of E.S.R. (mm/hrs) values pooled over time-interval on exposure to slightly higher concentration of drugs showed decreasing trend in all treatment group over control group. All these values were non-significantly decreased by 0.05, 0.12 and 0.11 (mm/hrs) in Zerokeet, Deltamethrin and Cypermethrin treatment group respectively.

Due to slightly higher dose of different drugs at 98-hrs. the E.S.R. value again decreased by 0.010 over 0-hour. But this change was also found to be non-significant.

The analysis of variance of the effect of different

drugs on slightly higher concentration on E.S.R. values at 0-hours to 98-hours have been depicted in **Table 49**. The ANOVA revealed non-significant difference between time-interval, drugs and interaction between drugs.

Table - 48 Mean, S.E. and C.V. % of E.S.R. (m.m./hour) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean
	<u>0-hrs</u> Mean ±S.E.	C.V.%	<u>98-hrs</u> Mean ±S.E.	C.V.%	
Cypermethrin	5.52 ±0.27	11.05	5.32 ±0.27	11.27	5.42 ^a
Deltamethrin	5.44 ±0.24	9.92	5.38 ±0.35	14.49	5.41 ^a
Zerokeet	5.46 ±0.27	11.17	5.5 ±0.20	8.18	5.48 ^a
Control	5.44 ±0.31	12.86	5.62 ±0.35	13.87	5.53 ^a
Overall Mean	5.465 ^a		5.455 ^a		

N.B. : Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

Table - 49 Analysis of variance of E.S.R. (m.m./hr) under different acaricides (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	0.03	0.07 N.S.
Between Time Interval	1	0.09	0.23 N.S.
Error	35	0.38	

N.B. = N.S. Non-Significant

Effects of Acaricides on Packed cell volume (P.C.V. %) volume on recommended dose concentration

The mean, S.E. and C.V. % of P.C.V. % under recommended concentration of Acaricides at 0-hrs (pretreatment) and at 98-hrs (post treatment) are presented in **Table 51**. The overall mean values were ranged between 40.0% to 41.0%. The values observed on treatment with Cypermethrin, Deltamethrin and Zerokeet group decreased over control group by 0.1%, 0.9%, 0.6% respectively. But all these differences were found to be non-significant from control. However, there was no significant differences ($p < 0.05$) noted between post treatment (98-hrs) and pretreatment (0-hrs) observations.

Table - 50 Mean, S.E. and C.V. % of Packed Cell Volume (%) under different Acaricides (Recommended dose) at different time-intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean
	0-hrs		98-hrs		
	Mean ±S.E.	C.V.%	Mean ±S.E.	C.V.%	
Cypermethrin	41.4 ±0.6	3.2	40.2 ±0.37	2.06	40.8 ^a
Deltamethrin	40.8 ±0.37	2.04	39.2 ±0.58	3.31	40.0 ^a
Zerokeet	40.6 ±0.4	2.20	40 ±0.28	1.57	40.3 ^a
Control	41.2 ±0.58	3.16	40.6 ±0.6	3.30	40.9 ^a
Overall Mean	41.0 ^a		40.0 ^b		

N.B. : Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

The analysis of variance on the effect of different drugs at recommended concentration on P.C.V. % at 0 and 98-hrs have been depicted in **Table 52** which reveals

significant differences between time-intervals and non-significant difference between drugs and interaction between drugs.

Table - 51 Analysis of variance of Packed Cell Volume (%) under different acaricides (Recommended dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	1.8	1.55 N.S.
Between Time Interval	1	10	8.62*.
Error	35	1.16	

N.B. : (1) N.S. - Non-Significant
(2) Significant at $p < 0.05$

Effect of acaricides on Packed Cell Volume (P.C.V. %) on slightly higher dose concentration:-

The mean, S.E. and C.V. % of Packed Cell Volume (%) at slightly higher concentration on spraying of different drugs are presented in **Table 52**. The overall mean values were ranged between 39.9% to 40.9%. The values observed on treatment with Cypermethrin, Deltamethrin and Zerokeet group decreased over control group by 0.1%, 1%, 0.4% respectively. But all these differences were found to be non-significant from control. There was no significant differences noted between post treatment (98 hrs.) and pretreatment (0-hrs) observations.

Table - 52 Mean, S.E. and C.V. % of Packed Cell Volume (%) under different Acaricides (Slightly higher dose) at different time intervals in dogs

Name of Acaricides	Time Intervals				Overall Mean
	0-hrs Mean ±S.E.	C.V.%	98-hrs Mean ±S.E.	C.V.%	
Cypermethrin	41.4 ±0.50	2.75	40.2 ±0.37	2.06	40.8 ^a
Deltamethrin	40.2 ±0.2	1.11	39.6 ±0.50	2.87	39.9 ^a
Zerokeet	40.4 ±0.24	1.35	40.6 ±0.24	1.33	40.5 ^a
Control	41.0 ±0.44	2.43	40.8 ±0.37	2.04	40.9 ^a
Overall Mean	40.75 ^a		40.3 ^a		

N.B. : Overall mean with different superscripts differs significantly at $p < 0.05$ separately in row and column

Table - 53 Analysis of variance of Packed Cell Volume (%) under different acaricides (Slightly higher dose) at different time interval in dogs

Source of variation	D.F.	M.S.	F
Between Treatment	3	2.02	2.77 N.S.
Between Time Interval	1	2.03	2.78 N.S.
Error	35	0.73	

Significant at $p < 0.05$

The analysis of variance for the effect of drugs treatment on P.C.V. at two time intervals and between

treatment groups have been depicted in **Table 53**. Analysis of variance revealed non-significant differences of P.C.V. between time intervals and between treatments at their slight higher concentration. ✕

DISCUSSION

Ectoparasites found on dogs, primarily ticks, represent a threat to their health and well being. Tick infestation are of great importance in the production of diseases to its host. Apart from direct physical damage and unthriftiness ticks also transmit disease organism, blood loss, allergic responses and damage of skin. The chemical acaricides widely in use for control are not only toxic, hazardous to man and environment but also resistance inducing (Nolan 1987). Of known chemical acaricides, pyrethrin are supposed to be least toxic to mammals and have minimum deleterious effect on the environment. The synthetic pyrethrin are reported to be effective even against resistant inducing strain to ticks and long time protection against reinfestation. (Bullman *et. al.* 1981 and Stubb *et. al.* (1982)

Presently some herbal preparation were also evaluated and reported for effective control of ticks.

The present study therefore planned to evaluate the comparative efficacy of some widely used pyrethrin compound viz. Cypermethrin and Deltamethrin and a herbal preparation Zerokeet to conclude their marginal difference in their therapeutic efficacy on application of their recommended and slightly higher doses, longer residual effect and toxicity in the body of treated dogs on the basis of post treatment haematological and some physiological parameters which help in direction of safe use and optional choice among various conventional and non-conventional insecticide.

Perusal and available literature also indicate that no systematic work has been carried out to study the incidence of ticks in various breeds or in general dog population in Patna. Many ticks borne haemoprotezoan parasites like *Babesia canis*, *E. canis* are particularly very

common in local dog population. Hence various breeds and stray dogs population were screened out for identification of various tick species infesting the local dog population of Patna as in relation to their breed, season and age etc.

A total of 445 (67.22%) dogs were found positive for the infestation of various tick species out of total 662 samples examined. (Maximum infestation was noted in the stray, cross bred and spitz dogs (70.72%)). The environmental factors such as higher relative humidity and temperature, congenial condition of local dogs made them easily get infested with tick. Unhealthy and unhygienic conditions of stray dogs, hairy texture of spitz variety, improper and non practising of tick control measures and least care bestowed upon cross bred dogs leads to higher rate of infestation among these varieties where as proper management, care and monitoring is associated with somewhat (33-55%) lesser range of infestation among high quality breeds such as Altatian, Labrador and Doberman. All total positive dogs were infested with brown dog ticks (*Rhipicephalus sanguineus*) which was also dominant in mixed infestation with *Haemophysalis bispisona* (5.16%). Any other species was not accounted through out the experimental period. *Haemophysalis bispisona* only present in the cases of mixed infestation, that too only in stray, cross bred and Altatian varieties.

Sinha *et. al.* (1987) observed higher incidence of tick infestation in stray dogs as compared to 92.8% of pet dogs. The most common tick species was *R. sanguineus* in local dogs population of Bihar. Dykstra *et. al.* (1997) reported that most frequent observed species was brown dog tick, 15 to 21 % of dogs in Texas where as Camacho *et. al.* (2003) found *Ixodes hexagonas* and *Ixodes ricinus* from dogs in Spain but again in Greece incidence (89.3%) of brown dog tick was predominant. Papazahoriadou *et al* (2003)

In concurrence of present result Miranpuri and

Singh (1978) also noted *H. intermedia* and *R. sanguineus* in dogs. Wilamowski *et. al.* (1999) also reported Brown dog tick was the most common tick in dog of Isreal and several cases of tick attachment to human being or handlers of dogs were also reported. Study revealed that despite of following various control measures, tick infestation is still widely prevalent throughout the local population of dogs and specially brown dog tick in dogs.

Highest incidence of tick infestation was noted in monsoon (July to October) followed by Summer (March to June) and least rate was in winter months (Nov. to Feb.) in present study throughout the population of pet and stray dogs. The study coincides with reports of Khan and Srivastava (1988) and Dykstra *et. al.* (1997) as they reported peak infestation from mid June to November followed by March to May; and between May to August respectively. But highest incidence in summer months reported by Sinha *et. al.* (1987), Mum cuoglu *et. al.* (1993), Inocuma *et. al.* (1995), Cruz *et. al.* (1999) however Sinha *et. al.* (1987) also reported least infestation in winter months.

The possible factors leading to highest rate of tick infestation during monsoon in present study may be suggested by higher rate of relative humidity, optimal temperature favourable for hatching of egg and despersion of developmental stages in pasture in this season which provide easy tick contact or transmission to dogs. Swampy, humid and improper cleaning of indoor areas provide better chances of maturation of tick stages and then contact of ticks to healthy animals even in indoor surroundings has a positive correlation with tick infestation as also reported by Cruz *et. al.* (1999) and Papazariadou *et. al.* (2003).

The present result also indicated that severity of tick infestation increased with advancement of age in all breeds of dogs. The study on influence of age on tick

infestation is very scanty but the reason suggested lowest infestation in newly born and young pups (0-3 months) may be that they are less hairy and continuously licked by their mother bitch throughout the body. Moreover in case of pet pups owners bestowed their best attention and care towards young pups and confinement in indoor area restrict them from getting heavy infestation. Whereas indiscriminate and non schedule acaricidal application of acaricide may also develop resistance in tick species in adult animals. No study however accounted by author on tick infestation as per the age group of dogs.

Study to evaluate comparative efficacy was carried out between pyrethroid compounds Cypermethrin and Deltamethrin and a herbal product Zerokeet against larval, nymph and adult stages of tick infestation on dogs, at their recommended concentration 1 ml/lit, 2 ml/ lit and 100/ 200 ml respectively and slightly higher concentration 1.25 ml/lit, 2.5 ml/lit and 125 ml/200 ml respectively.

The percent efficacy of these drugs compared in terms of no. of tick stage drop off and their residual period. The percent efficacy on drop off ticks was evaluated at 24 hours and 48 hours of post treatments following single spray of both concentration of each drug. Considering the control of tick as per the percent efficacy of acaricides at their recommended doses denotes that Cypermethrin, Zerokeet and Deltamethrin were 95-100%, 93-98% and 75-89% respectively effective against all stages of ticks within 24 hours of post treatment and at 48 hours almost 98-100% ticks of all stages were dropped off on all post treatment groups.

Similarly on spraying of Cypermethrin and Zerokeet at slightly higher concentration 100% control were seen against all stages of tick within 24 hours however percent efficacy indicates 90 to 98 % within various stages of ticks on spraying of Deltamethrin at 24 hours. 100 % eradication were noted within 48 hours on spraying of all acaricides

at their slight higher concentration. Result based upon percent efficacy of drugs indicated superior performance of Cypermethrin. But performance of recommended doses at 48 hrs and slightly higher doses indicate Zerokeet is also equally effective to Cypermethrin, however Deltamethrin is most frequent choice for tick control in dogs and other animals but present result indicated it has got slower effect than other two compounds. Study on residual effect based upon reinfestation of these acaricides also indicated that maximum effectiveness of Cypermethrin followed by Zerokeet and then Deltamethrin.

Present results are in line of finding of Singh (2000) who evaluated in a comparative trial, some chemical and herbal acaricides, indicated superior performance Cypermethrin and Zerokeet over Deltamethrin as per concerned tick mortality and residual effect. Srivastava *et. al.* (2001) also reported superiority of Cypermethrin over Deltamethrin but suggested that a resistance build up of these synthetic pyrethroid compound is high as these acaricides cannot be recommended at higher doses, because of presence of Cyclopropane ring produced immense irritation and photosensitisation. In present trial however there was no sign of irritation or any other side effect with ⁱⁿ observed ^{time} even on application of slightly higher doses ^{of} Cypermethrin or Deltamethrin and percentage of control was also higher within 24 hours but this can be suggested only after conducting trial (of slight higher doses) on large group of animals. Talukdar *et. al.* (1998) reported at 25 ppm and 500 ppm concentration of Deltamethrin had 72.86 to 100 % and 86.06 to 100 % respectively effectiveness against various stages of ticks. Residual effect was upto 18 - 20 days and 25 - 30 days respectively where as 25 ppm dilution of Deltamethrin caused complete recovery of ticks within 7 days as reported by Kinjavdekar and Parai (1995) with shorter residual effect and toxicity. Khan and Srivastava (1992) evaluated most effective synthetic Pyrethroid compound was Cyperkill followed by Permasect and Sumicidin and

period of reinfestation was 10 - 15 days. Tamang *et. al.* (1991) also reported various toxicity signs on application Cypermethrin in goats by drenching. Jani *et. al.* (1991) and Banerjee and Sangwan (1990) tried Butox (Deltamethrin) and reported that 0.0025% and 12.5 g/lit concentration respectively had cured rate of 83.34% against brown dog ticks on dogs on single application but with an interval of 7 days complete cure of tick was also reported. Endris *et. al.* (2002) indicated that a dose volume of 3 or 4 ml, 65% permethrin was needed to obtain adequate level and duration of ^{of efficacy} reinfestation against *R. sanguineus*. Results of present study very much similar to the finding of Srivastava *et. al.* (1993) as they reported 0.2% concentration of butox was found to be most effective against all stages of ticks within 48 hours. ^{repeat}

A herbal combination AV/EPP/14 which has similar constituents as Zerokeet (*Cerdrus deodara*, *Pongamia glabra*, *Azadirachita indica*, *Eucalyptus globulus* and *Acorus calamus*) had been evaluated by many workers. Kumar *et. al.* sprayed this preparation 1:4, 5 times at 6 day interval resulted 100% efficacy within 24 days and residual effect was found 30 days while Bhilegaonkar and Maske (1998) reported that this formulation at 1:4 was highly effective in killing larva, nymph and adult ticks (*R. sanguineus*) in 5, 30 and 72 hours respectively. Panda and Mishra (1997) reported that 1:0 to 1:4 concentration resulted 100% mortality of nymphal stages of *R. sanguineus* where as against adult females the drug resulted 100% mortality at 1:0 and 1:1 concentration, also slight adverse effect on hatchability of eggs and fecundity of females. Singh (1997) suggested requirement upto 3 applications at 5 days intervals of this drug. Sharma (1996) reported 1:4 formulation of AV/EPP/14 against tick in dogs on 2 to 3 applications for knocking off 50-60% of tick. Tripathy *et. al.* (1995) used 4 sprays of Ectozee - 50 (herbal formulation at 24 hrs intervals revealed 100% efficacy against ticks and reinfestation was noted only in 30% cases after 22 days. Maske and Bhilegaonkar

(1995) found Ectozee^{was} 25% and 100% effective in 8 hrs and 12 hrs respectively against nymphal and adult stages of ticks. The results and performance of Zerokeet 1:2 concentration in present trial (100% control of all stages within 48 hrs on single application) are also in line to these reports but repeat application has not been tried which may definitely will have longer residual effect. This drug was also found to be without any side effect but its quantity application is troublesome and hairy breeds may resulted with sometimes rough and dried hair, so a complete body wash with shampoo and conditioner are required for washing of not only herbal preparation but pyrethroid compound also. Some herbal shampoo (blaze) also reported effective for control and prevention of ectoparasites by Srivastava *et. al.* (1988).

There is very little information available in literature regarding the physiological and haematological changes occurring in animals due to application of pyrethroid compound or herbal product. However, various immunological, biochemical or haematological changes were ^{re}ported in infested dogs. *Rhipicephalus sanguineus* has a histamine blocking agent in salivary gland which provides an efficient mechanism for regulating the quantity of tissue fluid available for engorging tick (Chinery 1981). Infested dogs develop an immediate hypersensitivity reaction to *R. sanguineus* antigen and total serum IgE also increased significantly after infestation. Szabo *et. al.* (2003). Springell and O'Kelly (1971) reported metabolic changes like reduction in total body water, Hb %, plasma albumin and plasma cholesterol on tick infestation. Gabelhoff (1973) observed decrease in Hb content and adverse effect on erythropoiesis with increase in eosinophil and lymphocyte counts and decrease in neutrophils. Any acaricides which applied on host body also act or imbibe within the skin apart from causing the mortality of ticks. These chemical certainly cause some metabolic, neurological, haematological or physiological changes to host body on post application despite of various ^{proof} test

before commercialising these products, so in present trial some haematological and physiological changes were observed on post application Cypermethrin, Deltamethrin and Zerokeet at their recommended doses and slightly higher doses than recommended doses. All these changes were compared within treatment groups and within time intervals to not only decide the safer use of these products but also decide the marginal side effect caused by these products. The physiological parameters such as temperature, pulse and respiration rate were noted at 0, 6, 12, 24, 48 hours of post application where as haematological parameters were observed at 0, 24, 48, 72, 96 hours and P.C.V. and E.S.R. at 0 and 98 hours of post application.

TEMPERATURE

Analysis of variance undertaken in the present study showed that there was significant ($p < 0.05$) effect of both concentration of all the drugs as well as time interval on body temperature which started increasing within 6 hrs and onwards on post treatment in Cypermethrin and Deltamethrin treatment group both in recommended and slightly higher doses where as in Zerokeet treatment at recommended dose, it started increasing 12 hrs and onwards observed time intervals while at slightly higher dose it rised within 6 hrs and onwards, moreover temperature also started decreasing at 48 hrs. in slightly higher dose of Cyper and Deltamethrin group and Zerokeet group at 24 hrs.

A significant rise in body temperature may be due to stress caused by application of Deltamethrin was highest, followed by Cypermethrin & least affected group was Zerokeet on treatment with both recommended and slightly higher doses. However, a steep rise in temperature on slightly higher dose treatment was noted which started normalising spontaneously, so lower values were noted at 24 and 48 hours. Lawrance and Casida (1982) also

observed hyperthermic response i.e. significant rise in rectal temperature in pyrethroid treated rats and mice.

PULSE RATE

Significant rise ($p < 0.05$) in pulse rate was also noted in all treatment groups and between time intervals on treatment of both recommended and slightly higher concentration of drugs. Maximum pulse rate was increased in Deltamethrin treatment group on post treatment of both concentrations. The pulse rate rise up to 24 hrs. of post treatment but down trend was noted at 48 hrs. onwards post treatment of both trial. Rise in pulse rate was next in Cypermethrin treatment group and here also pulse rated higher upto 24 hrs. and then started decreasing at 12 hrs. on post treatment of both trial. The least affect noted in zerokeet treatment group of animals which also rised on treatments but started decreasing at 48 hrs. and 24 hrs. on recommended dose and slightly higher dose respectively. The observed data also revealed that at slightly higher doses treatment of drugs, more affected the pulse rate as it rapidly higher on all observed time-intervals. Lawrence and Casida (1982) also observed hyperthermic response i.e. significant rise in femoral pulse in pyrethroid treated rats and mice. Ray (1980) suggested that an exaggerated pulse may be due to release of catecholamine as a result of pyrethroid treatment on adrenal medulla and sympathetic fibres.

RESPIRATION RATE

Study on analysis of variance revealed significant change in respiration rate within all treatment groups and time-intervals. Maximum rise again noted in Deltamethrin treatment group on treatment with both doses upto 24 hours post treatment which observed decrease on 48 hours to its previous observation. The trend and values were followed by the treated groups with Cypermethrin. There was no significant difference in respiration rate

however found between control and Zerokeet treatment groups but slight rise in temperature were noted in Zerokeet group also, which started decreasing at 24 hours and 48 hours on post treatment of recommended dose and slightly higher dose respectively and non-significant changes were noted with the values of control. Lawrence and Casida (1982) and Ray (1980) reported rise in respiration rate due to release of Catecholamine on post treatment with pyrethroids. There was no report observed on post treatment changes on application of any herbal preparation.

HAEMOGLOBIN %

Ticks are virulent blood sucker. An adult female require 0.4 to 0.6 ml blood for its full engorgement. This loss of blood directly influence the Hb concentration. Significant reduction in Hb % was noted by Singh *et. al.* (1996) in tick infested dogs. The analysis of variance on post treatment of acaricides revealed that there was significant effect of acaricides treatments as well as time intervals on haemoglobin (gram %) in dogs infested with ticks. On treatment of recommended dose of Cypermethrin and Deltamethrin, Hb % further decreased upto 24 hours but again this started recovering from 48 hours and onwards observations with very slow rate. The reason may be suggested that the removal of ticks may affected this recovery. But animals on treatment with Zerokeet on recommended dose showed continuous fall throughout all the post treatment observed time periods and had no significant change from control which remain tick infested throughout the period study. This may attributed with long acting antiparasitic activity of herbal product to host body which stopped within 24 hours in pyrethroid treatment. On treatment of slightly higher dose with cypermethrin continuous fall in Hb % was noted in all observed period while it started rising at 48 hours in Deltamethrin group. Zerokeet also kept on decreasing throughout the period on slightly higher dose. But on the

basis of overall mean observation it was concluded that despite of continuous fall in Hb % in Zerokeet group (both concentration) the difference in decreased value was least in the this group (0-96 hrs) as comparing to pyrethroid compounds. Again the performance of Cypermethrin was next to Zerokeet and than Deltamethrin. Srivastava *et. al.* (2000) also reported reduction in the Haemoglobin value on treatment of Diazinon, deltamethrin at its recommended dose and ivermectin at slightly higher dose within 24 hours of spraying in cross bred calves infested with ticks. Springell and O'Kelly (1971) further reported decline in Hb% caused due to red cells consumed by the ticks and makeup of this loss is very difficult due to effect of tick toxin. The reports of Mishra *et. al.* (1996) are also in agreement with the findings of present study.

TOTAL ERYTHROCYTE COUNT

The analysis of variance revealed that there was significant effect of recommended dose and slightly higher dose of acaricides as well as the time interval on total erythrocyte count. The T.E.C. decrease upto 48 hours within treatment groups of recommended dose of cyper and deltamethrin and started recovering at later observations. On comparison with 0-hrs observation recovery in Deltamethrin group was better than cypermethrin where as in Zerokeet group continuous fall in T.E.C. value noted up to the period up to 72 hours but still the difference in overall mean was lesser than other two groups. While application on cypermethrin and deltamethrin (on treatment of slightly higher dose) recovery started with 48 hrs and onwards. Considering the difference from 0-hrs over 96 hrs., the improvement was much faster in Deltamethrin treatment group than Cypermethrin and the performance on recovery in zerokeet was superseded to these pyrethroid compounds. Reports of McCorkle *et. al.* (1980) also supported the post treatment evaluation on T.E.C value

of acaricides and also the present result and trend directly in line with the Hb% changes on posttreatment which is supposed to influence the T.E.C. value in same trend.

TOTAL LEUCOCYTES COUNT

The analysis of variance undertaken in the present study showed significant ($p < 0.05$) effect of recommended and slightly higher doses of acaricides, as well as time intervals on total leucocyte count (T.L.C.) in dog infested with ticks.

In all treatment group including control there was increase noted in values of T. L. Count but it was maximum in Cypermethrin group followed by Deltamethrin and zerokeet. There was a significant difference in the values of herbal and pyrethroid compounds both in treatment with recommended and slightly higher doses of acaricides. Winthrobed (1967) attributed leucocytosis in various toxic condition due to stimulating effect and release of catecholamine on post treatment of acaricides increases the count of leucocytes may be suggested for present result. Even tick biting also cause leucocytes infiltration around the blood vessels (Goksu and Orgencil 1970). McCorkle *et. al.* (1980) also noted increase in T.L.C. on treatment with Deltamethrin in chicken. Srivastava *et. al.* (2000) reported significant increase in T.L.C. value on treatment with various concentration of Deltamethrin and Diazinon in tick infested calves.

DIFFERENTIAL LEUCOCYTIC COUNT

Lymphocytes

Analysis of variance revealed that there was significant effect of treatment on lymphocyte count on treatment with recommended and slightly higher dose, maximum increase of lymphocyte noted in Cypermethrin group throughout the period of observation but in case of Deltamethrin increased value noted upto 24

hrs and then slightly decreased and then increasing trend was noted at 72 and 96 hours, minimum increase was also noted in Zerokeet group upto 48 hours and then started decreasing on treatment of recommended doses. Whereas on treatment with slightly higher concentration effect of treatment on lymphocyte count between time interval was found non-significant. Maximum increase noted in Cypermethrin treatment followed by Deltamethrin and Zerokeet. Particularly in Deltamethrin group the value at 48 hrs. even came less than pre-treatment values (0-hrs). Gabelhoff (1973) however noted increase in lymphocytes on tick infestation and further enhancement in present study on post-treatment may associated with increase of the T.L.C. values.

Neutrophils

Analysis of variance revealed significant effect of recommended dose of various treatments and time-intervals on neutrophil counts where as on post treatment with slightly higher dose, the effect between time interval was found non-significant. On Cypermethrin treatment with both doses a continuous decrease was noted which in this treatment group was maximum followed by same trend in Deltamethrin treatment groups. In Zerokeet group slight increase noted on recommended dose where as it remained almost same as pretreated values (0-hrs.) on treatment with Zerokeet slightly higher dose. Though Gabelhoff (1973) and Singh *et. al.* (1996) observed thorough infiltration of neutrophil in epidermis thickening on tick biting but neutrophil were generally reduced on infestation, any other study was not accounted on post treatment changes in neutrophil values.

Eosinophil

Non-significant difference in eosinophil values were noted on treatment of both doses of all three acaricides between treatments and time-intervals from control.

Gabelhoff (1973), Enokuna *et. al.* (1995) reported increase in number of eosinophils on tick infestation. The increase in plasma globulin on release of tick toxin was considered due to immunological allergic response which resulted increase in the count of eosinophil. It can be attributed in present trial also that all acaricides taken in the experiments definitely cause allergic responses to host body, therefore no significant changes were noted throughout the period of experiment over control or 0-hrs observation as there was pre-maintained eosinophilia due to tick toxins which continued on post treatment also.

Monocyte

Analysis of variance of present study showed significant ($P < 0.05$) effect in monocyte values due to recommended and slightly higher concentration of acaricides, however, there was no significant changes observed on treatment with recommended dose Cypermethrin and Zerokeet but it significantly decreased on treatment with recommended dose Deltamethrin. On treatment with slightly higher dose significant increase were noted within 48 hours and onwards in Cypermethrin group and decrease in Deltamethrin group on same time-intervals over all significant decrease also noted on Zerokeet slightly higher dose group values. It can be attributed that on application of slightly higher dose may affect the host body and cause immunological responses, toxicity chronicity against on various application of slightly higher doses as respective results observed during present trial.

P.C.V.

The analysis of variance revealed non-significant decrease in the value of P.C.V. between different treatment and time-intervals over control and 0-hrs readings respectively on application of recommended and slightly higher doses where as non-significant decrease was

noted in all treatment groups and between time intervals from control values. All these changes support the observation of decreasing T.E.C. value trend as T.E.C. has positive correlation with P.C.V. values.

E.S.R.

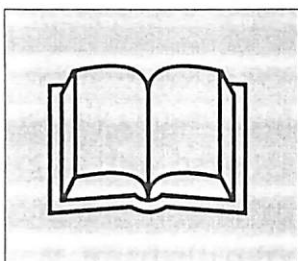
The analysis of variance revealed non-significant changes in value of E.S.R. between different time interval, treatments and within treatment over control. The chronicity impulsive by tick infestation remain unchanged on application of these acaricides.

Considering the above said changes in physiological and haematological parameters on application of recommended and slightly higher doses of acaricides, sizable proportion of metabolic changes occur in host body. While supportive reference on each particular changes is scanty, so further trial of slightly higher doses required in large group of animals and even extra time intervals. ✍

SUMMARY

As mentioned in dogs is a global problem. It is characterized by physical damage, metabolic changes, respiratory distress and changes to the host body. Most of the control measures are taken dependent upon the degree of severity which cause the respiratory distress and body with the action. It is not clear if the need for respiratory degree of the host body is truly revealed that the body is not yet in the way and that of proper application of conventional medicine.

CHAPTER - 5 SUMMARY AND CONCLUSION



and 120 ml/240 ml respectively and 1.25 ml/250 ml and 125 ml/250 ml respectively. On post treatment some physiological parameters temperature, pulse and respiration rate were assessed on 0-hrs (Pre-treatment), 6, 12, 24, 48 hours post treatments where as haematological parameters Hb %, T.B.C., T.L.C., D.L.C. were observed on 0-hrs (pre-treatment), 24, 48, 72, 96 hours post treatments P.C.V. and S.G.O.T. on 0 and 96 hours on post treatment of all three treatments. The two trial conducted separately each comprising of 20 naturally infected dogs for recommended 2.5 mg/kg

SUMMARY

Tick infestation in dogs is a global problem. It is characterized by physical damage, metabolic changes, transmitting toxin and diseases to the host body. Most of tick control measures are solely depend upon use of chemical acaricide which cause side effect, allergic responses and toxicity while their action, in contact with skin and tick mortality. Degree of tick infestation in locals area reveals that pet owners are still unaware of the ways and means of proper application of conventional acaricide which cause deleterious effect and accident to not only pets but also their handlers and surrounding environment.

Cypermethrin and Deltamethrin are now a days most widely chosen pyrithroid group of acaricides by pet owners as they have least side effect despite of having cyclopropane ring in structure and synergistic action. Zerokeet, a herbal bio-friendly product (*Cedurus deodara*, *Poncamia glabra*, *Azadirachita indica*, *Eucalyptus globulus*, *Acarus ealamus*) has been also reported effective against tick infestation. So, the present study was undertaken to evaluate the marginal difference in therapeutic efficacy of these acaricides on the basis of percent tick mortality and longer residual effect on the basis of period of reinfestation of ticks on application of Cypermethrin, Deltamethrin and Zerokeet on their recommended doses 1 ml/lit, 2 ml/lit and 100 ml/200 ml respectively and slightly higher doses 1.25 ml/ lit, 2.5 ml/lit and 125 ml/ 200 ml respectively upon randomly selected tick infested dogs. On post treatment some physiological parameters temperature, pulse and respiration rate were assessed on 0-hrs (Pre-treatment) 6, 12, 24, 48 hours post treatments where as haematological parameters Hb %, T.E.C., T.L.C., D.L.C. were observed on 0-hrs (pre-treatment), 24, 48, 72, 96 hours post treatments P.C.V. and E.S.R. on 0 and 96 hours on post treatment of all three acaricides. The two trial conducted separately each comprising of 20 naturally tick infested dogs for recommended and slightly

higher dose trial. The each group further divided into 4 sub-groups (5 dogs) for the application of Cypermethrin, Deltamethrin, Zerokeet and untreated control group. The observation were analyzed statistically between time-intervals and different treatments of drugs.

A brief study was on the incidence of various tick species in different breeds was also carried out to provide information of the degree tick infestation in local area and decide effective to and indiscriminate control schedule of acaricidal application in respect of breed, season and age of dogs.

The present study revealed that 67.22% dogs were found positive for tick infestation in different breeds of 662 pet and stray dogs in Patna and its surrounding area. Out of this, maximum (70-72%) infestation was noted between the stray, cross-bred and spitz varieties where as high quality breeds such as Altatian, Labrador and Doberman were carrying (33-35%) infestation rate, local environmental condition, unscheduled control practices, unhygienic and improper management and somewhat hairy texture leads to this higher rate of tick infestation in local breeds of dog. The present study also depicted the total positive samples were infested with brown dog ticks (*Rhipicephalus sanguineus*) which was also dominant in case of mixed infestation with *Haemophysalis bispinosa*.

The study on seasonal incidence revealed that highest rate of infestation was in monsoon (July to October) season followed by summer (march to June) and least rate was noted in winter months. Favourable condition like higher relative humidity, temperature favourable for egg hatching, maturation of developmental stages and easy transmission are the factors associated with higher rate of infestation in monsoon.

The study further indicated that severity of tick

infestation increases with advancement of age in all breeds of dogs. The reason suggested that being less hairy of newly born and young pups, and proper care taken towards these pups, keep them comparatively free from tick infestation.

The comparative evaluation of percent efficacy of Cypermethrin, Deltamethrin and Zerokeet against larva, nymph and adult stages of tick on their recommended doses resulted 95-100%, 75-89%, 93-98% respectively efficacy within 24 hours on post treatment and within 48 hours almost 98-100% ticks of all stages were dropped off within all treatment group. On slightly higher doses 100% efficacy evaluated on treatment with Cypermethrin and Zerokeet groups and nearly 90-98% tick dropped off in Deltamethrin group within 24 hours which also attained 100% efficacy within 48 hours. Reinfestation period of Cypermethrin group was also noted to be highest (25-32 days) followed by Zerokeet (20-30 days) and Deltamethrin (15-30 days) within both trials. The percent efficacy and period of reinfestation denotes the superior performance of Cypermethrin followed by Zerokeet and Deltamethrin on their recommended and slightly higher doses. The all experimental dogs were well tolerated even slightly higher doses of treatments without any visual side effects.

Table - 54 The effect on Physiological parameters (Overall mean) on use of (Recommended Dose) of acaricides

Physiological Parameters	<u>Recommended Dose (R.D.)</u>			Control
	Cyper-methrin	Delta-methrin	Zerokeet	
Temp. (F°)	102.18 ^a	102.55 ^b	101.59 ^c	101.40 ^{cd}
Pulse/ minute	87.36 ^a	91.2 ^b	81.48 ^c	81.6 ^{cd}
Resp. rate/minute	29.04 ^a	30.48 ^b	22.44 ^c	22.52 ^{cd}

	<u>Slightly Higher Dose (S. H. D.)</u>			
Temp. (F°)	102.55 ^a	103.07 ^b	101.51 ^c	101.73 ^{cd}
Pulse/ minute	88.84 ^a	91.5 ^a	83.32 ^b	83.5 ^{bc}
Resp. rate/minute	32.6 ^a	33.16 ^a	22.64 ^b	22.08 ^{bc}

Table - 55 The effect on Haematological parameters (Overall mean) on use of acaricides

	<u>Recommended Dose (R.D.)</u>			
Physiological Parameters	Cyper-methrin	Delta-methrin	Zerokeet	Control
Hb (%)	10.58 ^a	10.47 ^a	10.33 ^b	11.55 ^{bc}
TEC (10 ⁶ Xmm ³)	5.508 ^a	5.48 ^a	5.84 ^b	5.844 ^{bc}
TLC (10 ³ Xmm ³)	14.23 ^a	14.024 ^a	13.116 ^b	13.116 ^{bc}
Lymphocyte (%)	28.95 ^a (23.48)	28.64 ^{ab} (23)	28.25 ^b (22.44)	27.82 ^{bc} (21.96)
Neutrophil (%)	52.87 ^a (63.52)	54.11 ^b (65.64)	54.30 ^{bc} (65.96)	54.21 ^{bcd} (65.76)
Eosinophil (%)	18.33 ^a (9.92)	18.14 ^a (9.7)	17.67 ^a (9.24)	18.25 ^a (9.84)
Monocyte (%)	8.45 ^a (2.2)	7.17 ^b (1.6)	8.45 ^a (2.2)	8.47 ^a (2.24)
P.C.V. (%)	40.8 ^a	40 ^a	40.3 ^a	40.9 ^a
E.S.R. (mm/hrs)	5.93 ^a	5.86 ^a	5.65 ^a	5.59 ^a

	<u>Slightly Higher Dose (S. H. D.)</u>			
Physiological Parameters	Cyper-methrin	Delta-methrin	Zerokeet	Control
Hb (%)	9.78 ^a	10.90 ^b	11.24 ^{bc}	11.32 ^{cd}
TEC (10 ⁶ Xmm ³)	5.212 ^a	5.14 ^a	5.724 ^b	5.66 ^{bc}
TLC (10 ³ Xmm ³)	15.052 ^a	14.884 ^a	13.088 ^b	13.076 ^{bc}
Lymphocyte (%)	29.52 ^a (24.32)	28.53 ^b (22.84)	27.51 ^c (22.36)	28.07 ^{bd} (22.16)
Neutrophil (%)	52.63 ^a (63.14)	54.02 ^{bc} (65.48)	55.03 ^c (67.16)	54.16 ^{bcd} (65.72)
Eosinophil (%)	18.07 ^a (9.56)	18.40 ^a (10)	18.30 ^a (9.88)	18.23 ^a (9.84)

Monocyte (%)	9.16 ^a (2.56)	7.05 ^b (1.56)	6.96 ^c (1.52)	8.55 ^a (2.24)
P.C.V. (%)	40.8 ^a	39.9 ^a	40.5 ^a	40.9 ^a
E.S.R. (mm/hrs)	5.42 ^a	5.41 ^a	5.48 ^a	5.53 ^a

N. B. 1. Mean with different superscripts differ significantly row wise ($p < 0.05$)

2. The values of Lymphocyte, Neutrophil, Eosinophil and Monocyte are indicated the Arc sin value of corresponding means (Figure in parenthesis are original mean value)

Considering the overall mean between treatments the physiological parameters indicated exaggerated temperature, pulse and respiration rate in all treatment groups which was maximum in Cypermethrin group followed by Deltamethrin treatment where as non-significant changes were noted within Zerokeet group on application of both recommended and slightly higher doses. The effect on temperature, pulse and respiration rate of slightly higher did not vary much than recommended doses.

Haematological parameters viz. Haemoglobin percent, Total erythrocyte count decreased significantly on treatment with Cypermethrin and Deltamethrin on recommended and slightly higher doses. The maximum decrease of Hb % found in Cypermethrin group followed by Deltamethrin group in both trials but T.E.C. maximum decreased in Deltamethrin group and Cypermethrin group on treatment with recommended and slightly higher dose respectively. Reduction in Hb% and TEC count were also noted in Zerokeet group but it was found non-significant over control group.

Leucocytosis is the marked feature of tick infestation which further increased on treatment with pyrethroid compounds and was found maximum on treatment with Cypermethrin followed by Deltamethrin treatment in both trials of recommended and slightly higher doses, However,

changes were found non-significant on treatment with Zerokeet in both trials.

The study and evaluation of differential leucocyte count was carried out on basis of arc sin values corresponding to original mean.

The observation on lymphocyte count indicated significant increase in all treatment groups of recommended doses it was found maximum in Cypermethrin group and non-significant difference within Deltamethrin and Zerokeet treatment groups. However, on treatment with slightly higher doses maximum increase again noted in Cypermethrin group while it was non-significant in Deltamethrin over control group. A significant decrease was noted in lymphocyte value in Zerokeet slightly higher dose group over control.

Non-significant changes in Neutrophil count noted in Deltamethrin and Zerokeet group over control group in both trials while it significantly decreased and increased on treatment with recommended and slightly higher doses of Cypermethrin respectively.

Non-significant changes in monocyte count within Cypermethrin and Zerokeet group on treatment of recommended doses were ~~noted~~ which was found significantly decreased in Deltamethrin group. Significant decrease however noted on treatment with slightly higher doses which was maximum in Zerokeet group followed by Deltamethrin group but significant increases were noted in monocyte values in Cypermethrin group.

There was no significant changes occur in the P.C.V. count however decreased PCV was noted in all treatment groups following the trend of TEC count.

There was no significant changes in ESR value were noted on treatment with recommended and slightly

higher dose of all treatment. The values indicated that chronicity of infection were maintained even after treatments.

The study concluded that marked rise in temperature, pulse rate, respiration rate and lymphocytes occurred on treatment of acaricides at their recommended and slightly higher doses where as marked decrease was counted in the values of Hb %, TEC, PCV, Neutrophil and monocyte counts. Eosinophilia and increased ESR persisted throughout experimental period without any significant change in different treatment groups.

Despite of some enhanced value in physiological parameters observed on treatment with slightly higher dose, there was not very unusual trend marked comparing to recommended dose. The difference in values were also very minor which hardly affects (well tolerated) the treated animal even with slightly higher doses. The trend of haematological parameters also depicted the same picture. But the use of slightly higher doses can be recommended only after conducting repeat trials on large group of animal and within more time-intervals.

Cypermethrin was found to be most efficacious but Zerokeet was having least affect on physiological and haematological parameters but its higher quantity requirement (1 : 2) and comparing higher prices may be bottleneck for its easy application. Deltamethrin (Butox) has been dominated present local market but has least efficacious within these drugs and with lesser side effect than cypermethrin treatment. ✍

CONCLUSION

1. Infestation of ticks was found widely prevalent among local general population in dogs in Patna more specially in stray, cross bred and hairy varieties of dogs.
2. Most of the dogs were found infested with *Rhipicephalus sanguineus*.
3. Incidence of tick infestation was found maximum in monsoon & summer seasons.
4. Cypermethrin (1 ml/ litre wash spray) was found to be the most efficacious for the elimination of all stages of ticks followed by Zerokeet (1:2) which was almost equally effective ^{as cypermethrin} at its recommended doses, while Deltamethrin, which has dominated the present market but its comparative efficacy against ticks was found less effective and time taking than previous products. The same trend was evaluated in slightly higher doses and period of reinfestation.
5. zerokeet (1:2) had least toxic effect on physiological and hematological parameters followed by Deltamethrin and Cypermethrin at their recommended and slightly higher concentration.
6. Slightly higher dose of cypermethrin (1.25ml/lit.) and deltamethrin (2.5ml/lit.) casts considerable affect on physiological and hematological parameters in dogs but these changes were tolerated by the host within observed time intervals, so responses to these doses require to be tested in further trial cautiously in large group of animals and also by evluating other biochemical parameters.
7. Though zerokeet has no side effect even on its slightly higher doses (1.25:2) but its higher prices and uneasy application may be limiting factor than other two acaricides. ✍

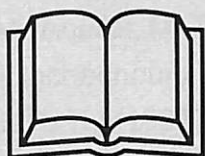
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APPENDIX

Plate-1 Male (Adult) *Rhipicephalus sanguineus* (10x10x)
Female (Adult engorged) *Rhipicephalus sanguineus*
(10x10x)

Plate- 2 Female (Adult engorged) *Rhipicephalus sanguineus*
(10x10x)

