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Comparative efficacy of Nemocid, Albomar and Alfanil in cases of spontaneous Toxocariasis at dog ward, teaching veterinary clinical complex, WBUAFS, Kolkata, West Bengal, India

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Abstract
Introduction: Toxocariasis is a worldwide parasitic infection and dogs serve as the most important definitive hosts for *Toxocara canis*: a causative agent of human toxocariasis and one of the most widespread zoonotic helminths worldwide.
Materials and methods: For this study out of 24/42 positive pups irrespective of age, sex and breeds were randomly selected. Total 30 pups divided into 5 groups of 6 puppies as: Group 1: Healthy control, Group 2: Untreated control, Group 3: Nemocid (Pyrantel pamoate) suspension @50 mg/kg bw/d orally as single dose, Group 4: Albomar (Albendazole) suspension @50 mg/kg bw/d for 3 days orally and, Group 5: Alfanil tablet (combination of Praziquantel, Pyrantel embonate and Oxantel embonate) @1 tab/10 kg bw/d orally as single dose. Animals were under observation for 9-months from treatment dosing and tests for clinical, haematological, biochemical and faecal examinations.
Results: Overall incidence of *Toxocara canis* infection in and around Kolkata city was 38.18%, with higher incidence during the monsoon (64.29%), sex-wise in males (66.67%), incidence-wise in age group 1-3 months, breed-wise in non-descript puppies (61.90%), and a decrease of haematological and biochemical parameters was observed in the pre-treatment infected pups. All three anthelmintics were found effective against the *Toxocara spp.* infection in the pups.
Conclusion: Considering the best efficacy, the combination of Praziquantel, Pyrantel embonate and Oxantel embonate at a dose rate of 1 tab/10 kg b.wt. as single dose can suitably be recommended for treatment of toxocariosis in pups.

Keywords: toxocariosis, *Toxocara canis*, zoonosis, efficacy, incidence, prevalence

1. Introduction
The name "*Toxocara*" is derived from the Greek word "toxon," meaning bow or quiver, and the Latin word "caro," meaning flesh (Bassert and Thomas, 2014) [1]. Toxocariasis is a worldwide parasitic infection, primarily rendered by *Toxocara canis* in dogs, *Toxocara cati* in cats and foxes and *Toxascaris leonina* in a wide range of carnivores (Chen *et al*., 2012) [2]. Dogs serve as the most important definitive hosts for *Toxocara canis*: a causative agent of human toxocariasis and one of the most widespread zoonotic helminths worldwide (Rostami *et al*., 2020) [3].
Toxocariasis in dogs is caused by ingestion of fully embryonated eggs or ingestion of infective larvae together with paratenic host of nematode roundworm *Toxocara canis* (Mondal and Basak, 2001) [4]. It is the most common *Toxocara* parasite of concern to humans, which puppies usually contract from their mother before birth or from her milk. In 3 to 4 weeks old pups, the larvae mature in the intestine and the worms start to produce large quantity of eggs which ultimately shed in to the environment through the faeces. Within 2 to 4 weeks the infective larvae can develop into eggs in the environment and the ingestion of these eggs can make one infected with toxocariasis (Centre for Disease Control, 2023) [5]. In an epidemiological perspective, animal hosts parasitized by adult worms in their gut can disseminate infection by shedding parasite eggs into environment (Eslahi *et al*., 2020) [6].
Epidemiological surveys have indicated that the prevalence of *Toxocara canis* in dogs ranged from 5.5% to 64.7% (Minnaar *et al*., 2002; Oliveira-Sequeira *et al*., 2002; Habluetzel *et al*., 2003; Rubel *et al*., 2003; Wang *et al*., 2006; Dai *et al*., 2009) [7-12]. Recent meta-analysis study has estimated overall prevalence of soil contamination with *Toxocara spp*. eggs in public places as 11.26% (95% CI 07.59–15.54%) across India (Bhangale *et al*., 2021) [13].

Toxocariasis due to several species of *Toxocara* and/or Toxascaris roundworms is still a seriously notifiable public health issue, particularly due to its intricate transmission routes (Eslahi *et al*., 2020) [6].

Normally a normal sized dog lightly infected with *Toxocara canis* passes 136 gm of faeces per day having 10000 Eggs Per Gram of Faeces (EPGF) (Ahmad *et al*., 2011) [14]. The estimation of EPGF is very simple yet effective method to predict the severity and parasitic burden of *Toxocara canis* in dogs (Shulaw, 2011) [15]. After administration of anthelmentics, a reduction has been noticed in the number of ova discharged in faeces and improvement in haemato-biochemical parameters and, the efficacy can be compared by relative decrease in EPGF and also relative improvement in haemato-biochemical parameters (Rao and Suryanarayana, 1995) [16].

2. Materials and Methods

2.1 Study design

For this study a total of 110 puppies aged 0-6 months, irrespective of sex, breed and size and brought to the Dog ward under Teaching Veterinary Clinical Complex (TVCC), Faculty of Veterinary and Animal Sciences under West Bengal University of Animal and Fishery Sciences, Kolkata, India were examined for spontaneous ascariasis infection by *Toxocara canis*.

Out of 110 pups tested, 42 were found to be positive for ascariasis of which 24 number of pups were selected at random. Total 30 pups divided into 5 groups of 6 puppies each were divided as: Group 1: Healthy control, Group 2: Untreated control, Group 3: Nemocid (Pyrantel pamoate) suspension @50 mg/kg bw/d orally as single dose, Group 4: Albomar (Albendazole) suspension @50 mg/kg bw/d for 3 days orally and, Group 5: Alfanil tablet (combination of Praziquantel, Pyrantel embonate and Oxantel embonate) @1 tab/10 kg bw/d orally as single dose. Animals under study group (n = 30) were under observation for 9-months from treatment dosing and tests for clinical, haematological, biochemical and faecal examinations were carried out once a month. The study was carried with approval of Institutional Animal Ethics Committee, West Bengal University of Animal and Fishery Sciences, Kolkata-37, India.

2.2 Prevalence of Toxocariosis

Faecal samples were collected from different portions of faecal volume. The samples were kept in individually marked vials and no preservative was added. For qualitative examination, samples were examined by direct smear method and centrifugation technique, and quantitatively by modified Mc. Master's Technique (Soulsby, 1982) [17] for determining the eggs per gram (EPG) of faeces. Incidence was calculated based on observations specific to breed, age, sex and season and overall incidence of *Toxocara spp*. infection in pups was calculated as:

$$\text{Overall indicence of Toxocara spp.} = \frac{\text{No. of positive samples for Toxocara spp.}}{\text{Total no. of samples}} \times 100$$

2.3 Haemato-biochemical analysis

2.3.1 Blood sample collection and processing: The blood samples were collected from all the group of pups on 0th, 7th, 15th and 30th days for the haematological and biochemical studies. About 7 mL of blood was collected aseptically from the cephalic or saphenous veins with a sterile 10 mL disposable syringe from each animal. Out of 7 mL, 1 mL was transferred from the syringe to the vials containing EDTA for estimation of Haemoglobin (Hb), Differential Leukocytic Count (DLC), Total Erythrocytic Count (TEC) and Total Leukocytic Count (TLC). 4 mL of blood was allowed to clot to collect serum and stored at -20° C after proper levelling for biochemical investigations. Another 2 mL of blood was transferred to the serum vials containing heparin for estimation of Glucose.

2.3.2 Estimation of haematological parameters: Hb (g/dL) was estimated by Sahli's method (Schalm *et al*., 1975) [18]. TEC (x10^6/µL) and TLC (x10^3/µL) of blood was estimated by the Haemocytometer. DLC (%) was estimated by hematocrit tube method and blood films stained by Giemsa staining method (Schalm *et al*., 1975) [18].

2.3.3 Estimation of serum biochemical parameters: TSP (g/dL) was estimated spectrophotometrically by Modified Biuret End Point Assay methods (Koller, 1984) [19]. Serum albumin (g/dL) was estimated spectrophotometrically by Bromocresol Green End Point Assay method (Corcoran and Durran, 1977) [20] and serum globulin (g/dL) was calculated by subtracting the serum albumin value from the total serum protein values. The blood glucose level (mg/dL) was estimated spectrophotometrically by GOD-POD based end-point method (Tietz, 1976) [21]. Serum AST (IU/L) and serum ALT (IU/L) values were estimated spectrophotometrically by 2,4- DNPH method (Reitman and Frankel, 1957) [22]. Total serum bilirubin was estimated spectrophotometrically by Malloy and Evelyn Method (Malloy and Evelyn, 1937) [23] and Serum creatinine level was estimated spectrophotometrically by Alkaline Picrate method (Toro and Ackermann, 1975) [24]. All estimations were carried out by using commercial kits supplied by Span Diagnostic Ltd., Surat, India.

2.4 Therapeutic trail: Therapeutic trial was conducted to evaluate the efficacy of three different anthelmintic drugs for treatment of the naturally infected *Toxocara spp*. infection in pups in different groups to evaluate their efficacies. For assessing the reduction of EPG count, the faecal samples were collected on day '0' before treatment and subsequently on 3rd, 7th, 14th and 28th days of post medication. The percent efficacy was done as follows (Soulsby, 1994) [25]:

$$\text{Efficacy \%} = \frac{\text{EPG before treatment} - \text{EPG after treatment}}{\text{EPG before treatment}} \times 100$$

2.5 Statistical analysis: Data obtained under present investigation were entered in spreadsheet for descriptive analysis (mean and percent) and SPSS version 10.0 was used for ANOVA (Snedecor and Cochran, 1976) [26].

3. Results and Discussion

3.1 Incidence

Overall incidence of *Toxocara spp*. infection in pups in this study was found 38.18% and this finding corroborates with the findings of Negrea (2005) [27] who also reported that, other than transplacental infection, puppies are mostly infected from the mother's faeces.

$$\text{Overall indicence of Toxocara spp.} = \frac{42}{110} \times 100 = 38.18\%$$

Higher incidences of *Toxocara canis* infection was recorded in males (66.67%), monsoon season (64.29%), age group of 1-3 months (54.77%), and amongst the non-descript pups (61.90%) [Table 1]. Season wise incidence might be due to effect of rainfall in the translocation and acquisition of L3 infective larvae in the environment as referred to by Soulsby (1994) [25].

3.2 Haematological parameters: The results of the haematological observations in different groups of pups have been presented in Table 2 and Figure 1. Hb levels of the infected pups of Groups 2, 3 4 and 5 were significantly (p<0.01) lower than Group 1 on 0th day simulated with the observations of Chattha *et al.* (2009) [28] in pups with toxocariosis which might be due to extensive tissue damage of the intestinal wall and blood sucking behaviour of the infective larvae of *Toxocara spp.* in puppies as also opined by Deger *et al.* (1997) [29]. The TEC of the infected puppies of Groups 2, 3, 4 & 5 also showed significant (p<0.01) declination on 0th day than Group 1, but following treatment in Groups 3,4 & 5, the values improved markedly on 30th day. The TLC of the infected puppies (Groups 2, 3, 4 & 5) increased significantly (p<0.01) on 0th day than the healthy control puppies (Gr.1) indicating marked leucocytosis and are in conformity with Sharma *et al.* (2010) [33] who also noted increased TLC with toxocariosis in pups.

Table 1: Distribution of positive samples of *Toxocara canis* in pups

Age-wise incidence			Breed-wise incidence			Season-wise incidence		
Age	**Cases**	**Incidence**	**Breed**	**Cases**	**Incidence**	**Season**	**Cases**	**Incidence**
1-3 months	23	54.77%	Labrador	1	2.38%	Winter (Dec-Feb)	4	9.52%
4-6 months	19	45.23%	German Spitz	4	9.52%	Summer (Mar-May)	11	26.19%
Sex-wise incidence			German Shepherd	1	2.38%	Monsoon (Jun-Aug)	27	64.29%
Sex	Cases	Incidence	Golden Retriever	1	2.38%	Total samples	Positive samples	Negative samples
Male	28	66.67%	Cross-breed	9	21.44%	110	42	68
Female	16	33.33%	Non-descript	26	61.90%	Overall Incidence		38.18%

There was a significant (p<0.01) elevation of Neutrophil percentages in Groups 2, 3, 4 & 5 than the Group 1 puppies and simulated with the observations of Sharma *et al.* (2010) [33]. There was further increase of the values in Group 2 puppies, while declinations were noted in Groups 3, 4 & 5. In treatment groups, a significant improvement (p<0.01) in lymphocyte percentage observed in Group 3 on 30th day, and on 15th day in Group 4 and Group 5. This improvement in lymphocytic percentages indicated the control of secondary bacterial infections in toxocariosis.

The eosinophil levels were significantly higher (p<0.01) in Groups 2, 3, 4 and 5 compared to the Group 1 healthy control on 0th day and is in agreement with the observations of Chatterjee *et al.* (1993) [31], Ogunkoya *et al.* (2006) [32], Sharma *et al.* (2010) [33] and Devi *et al.* (2011) [34], who also noted eosinophilia in *Toxocara* infections. Chatterjee *et al.* (1993) [31] opined that eosinophilia could be due to the pro-inflammatory process and tissue injuries triggered following larval migration and subsequent release of histamine or serotonine like chemo attractants in dogs. The monocyte levels of the healthy pups in Group 1 corroborates with the observations of Coles (1986) [35], and a decrease level of monocytes on 0th day corroborates with the findings of Sharma *et al.* (2010) [33] who observed a decrease in monocyte levels in experimentally infected chickens with *Toxocara spp.*

Table 2: Haematological changes in control and treatment groups of pups

Sl. No.	Parameter	Group	Pre-treatment	Days of observation post-treatment		
			0th Day	7th Day	15th Day	30th Day
1	Hb (g/dL)	I	13.89 ± 0.10^{a}	13.93 ± 0.11^{a}	13.95 ± 0.10^{a}	13.96 ± 0.10^{a}
		II	$9.18\pm0.08^{d}_{w}$	$9.02\pm0.05^{e}_{wx}$	$8.87\pm0.06^{d}_{xy}$	$8.77\pm0.05^{d}_{y}$
		III	$9.40\pm0.03^{cd}_{z}$	$9.69\pm0.02^{d}_{y}$	$10.35\pm0.11^{c}_{x}$	$11.25\pm0.03^{bc}_{w}$
		IV	$9.74\pm0.06^{b}_{y}$	$10.35\pm0.05^{b}_{x}$	$11.02\pm0.04^{b}_{w}$	$11.19\pm0.06^{c}_{w}$
		V	$9.54\pm0.03^{bc}_{z}$	$9.96\pm0.04^{c}_{y}$	$10.51\pm0.04^{c}_{x}$	$11.48\pm0.04^{b}_{w}$
2	TEC (x10^6/µL)	I	$7.02\pm0.02^{a}_{y}$	$7.05\pm0.01^{a}_{xy}$	$7.08\pm0.01^{a}_{wx}$	$7.12\pm0.01^{a}_{w}$
		II	$5.07\pm0.04^{b}_{w}$	$4.85\pm0.03^{b}_{x}$	$4.77\pm0.02^{c}_{xy}$	$4.73\pm0.02^{d}_{y}$
		III	$4.82\pm0.03^{c}_{x}$	$4.89\pm0.02^{b}_{x}$	$4.94\pm0.04^{b}_{x}$	$5.18\pm0.02^{b}_{w}$
		IV	$4.60\pm0.02^{d}_{z}$	$4.70\pm0.01^{c}_{y}$	$4.83\pm0.01^{c}_{x}$	$4.74\pm0.02^{c}_{w}$
		V	$4.44\pm0.01^{c}_{z}$	$4.56\pm0.02^{d}_{y}$	$4.74\pm0.02^{c}_{x}$	$5.11\pm0.02^{b}_{w}$
3	TLC (x10^3/µL)	I	11.39 ± 0.07^{b}	11.47 ± 0.08^{b}	11.54 ± 0.07^{c}	11.63 ± 0.08^{cd}
		II	14.35 ± 0.30^{a}	14.47 ± 0.29^{a}	14.57 ± 0.30^{ab}	14.67 ± 0.30^{a}
		III	13.78 ± 0.33^{a}	13.63 ± 0.32^{a}	13.42 ± 0.33^{ab}	13.22 ± 0.33^{b}
		IV	14.31 ± 0.43^{a}	14.08 ± 0.43^{a}	13.78 ± 0.42^{ab}	13.13 ± 0.35^{ab}
		V	13.97 ± 0.33^{a}	13.65 ± 0.34^{a}	13.13 ± 0.35^{b}	12.69 ± 0.35^{bc}
4	Neutrophil (%)	I	$70.58\pm0.20^{b}_{x}$	$71.66\pm0.21^{b}_{w}$	$70.17\pm0.17^{c}_{xy}$	$69.50\pm0.22^{c}_{y}$
		II	$73.83\pm0.30^{a}_{x}$	$74.66\pm0.42^{a}_{wx}$	$75.50\pm0.42^{a}_{wx}$	$76.167\pm0.60^{a}_{w}$
		III	74.00 ± 0.57^{a}	73.66 ± 0.61^{ab}	72.83 ± 0.61^{b}	71.83 ± 0.54^{b}
		IV	$74.33\pm0.61^{a}_{w}$	$73.17\pm0.65^{ab}_{wx}$	$72.00\pm0.65^{bc}_{x}$	$68.67\pm0.21^{c}_{y}$
		V	$75.00\pm0.68^{a}_{w}$	$73.17\pm0.75^{ab}_{wx}$	$71.17\pm0.75^{bc}_{x}$	$68.17\pm0.54^{c}_{y}$
5	Lymphocyte (%)	I	$23.08\pm0.27^{a}_{xy}$	$22.33\pm0.21^{a}_{y}$	$23.83\pm0.31^{a}_{wx}$	$24.16\pm0.16^{a}_{w}$
		II	18.33 ± 0.56^{b}	17.66 ± 0.61^{b}	17.16 ± 0.60^{d}	16.83 ± 0.40^{c}
		III	$17.50\pm0.56^{b}_{x}$	$18.33\pm0.42^{b}_{wx}$	$19.00\pm0.26^{cd}_{wx}$	$19.67\pm0.21^{b}_{w}$

		IV	$17.66\pm0.49^{b}_{y}$	$18.83\pm0.31^{b}_{y}$	$20.50\pm0.50^{bc}_{x}$	$24.33\pm0.33^{a}_{w}$
		V	$17.33\pm0.76^{b}_{x}$	$19.33\pm0.61^{b}_{x}$	$22.00\pm0.61^{ab}_{w}$	$24.33\pm0.56^{a}_{w}$
6	Eosinophil (%)	I	$3.00\pm0.00^{b}_{w}$	$2.83\pm0.01^{b}_{w}$	$2.66\pm0.21^{c}_{wx}$	$3.00\pm0.00^{b}_{w}$
		II	5.16 ± 0.54^{a}	5.50 ± 0.61^{a}	5.16 ± 0.60^{a}	5.66 ± 0.49^{a}
		III	$5.66\pm0.42^{a}_{w}$	$5.00\pm0.51^{a}_{wx}$	$4.66\pm0.33^{ab}_{wx}$	$3.83\pm0.16^{b}_{x}$
		IV	$5.33\pm0.49^{a}_{w}$	$4.66\pm0.21^{a}_{wx}$	$3.66\pm0.21^{bc}_{xy}$	$3.16\pm0.30^{b}_{y}$
		V	$5.66\pm0.42^{a}_{w}$	$4.50\pm0.22^{a}_{x}$	$3.66\pm0.21^{bc}_{xy}$	$3.00\pm0.00^{b}_{y}$
7	Monocyte (%)	I	3.33 ± 0.21^{a}	3.16 ± 0.17	3.33 ± 0.21^{a}	3.33 ± 0.21^{c}
		II	$2.67\pm0.21^{ab}_{w}$	$2.16\pm0.17_{w}$	$2.17\pm0.16^{b}_{w}$	$1.33\pm0.21^{d}_{x}$
		III	$2.83\pm0.17^{ab}_{x}$	$3.00\pm0.36_{x}$	$3.50\pm0.22^{a}_{x}$	$4.67\pm0.21^{a}_{w}$
		IV	$2.67\pm0.21^{ab}_{x}$	$3.33\pm0.33_{wx}$	$3.83\pm0.16^{a}_{w}$	$3.67\pm0.21^{bc}_{w}$
		V	$2.00\pm0.25^{ab}_{y}$	$3.00\pm0.36_{xy}$	$3.17\pm0.31^{a}_{x}$	$4.50\pm0.22^{ab}_{w}$

Mean bearing different superscripts (a, b, c, d and e) in a column differ significantly (p<0.01).
Mean bearing different subscripts (w, x, y and z) in a row differ significantly (p<0.01).

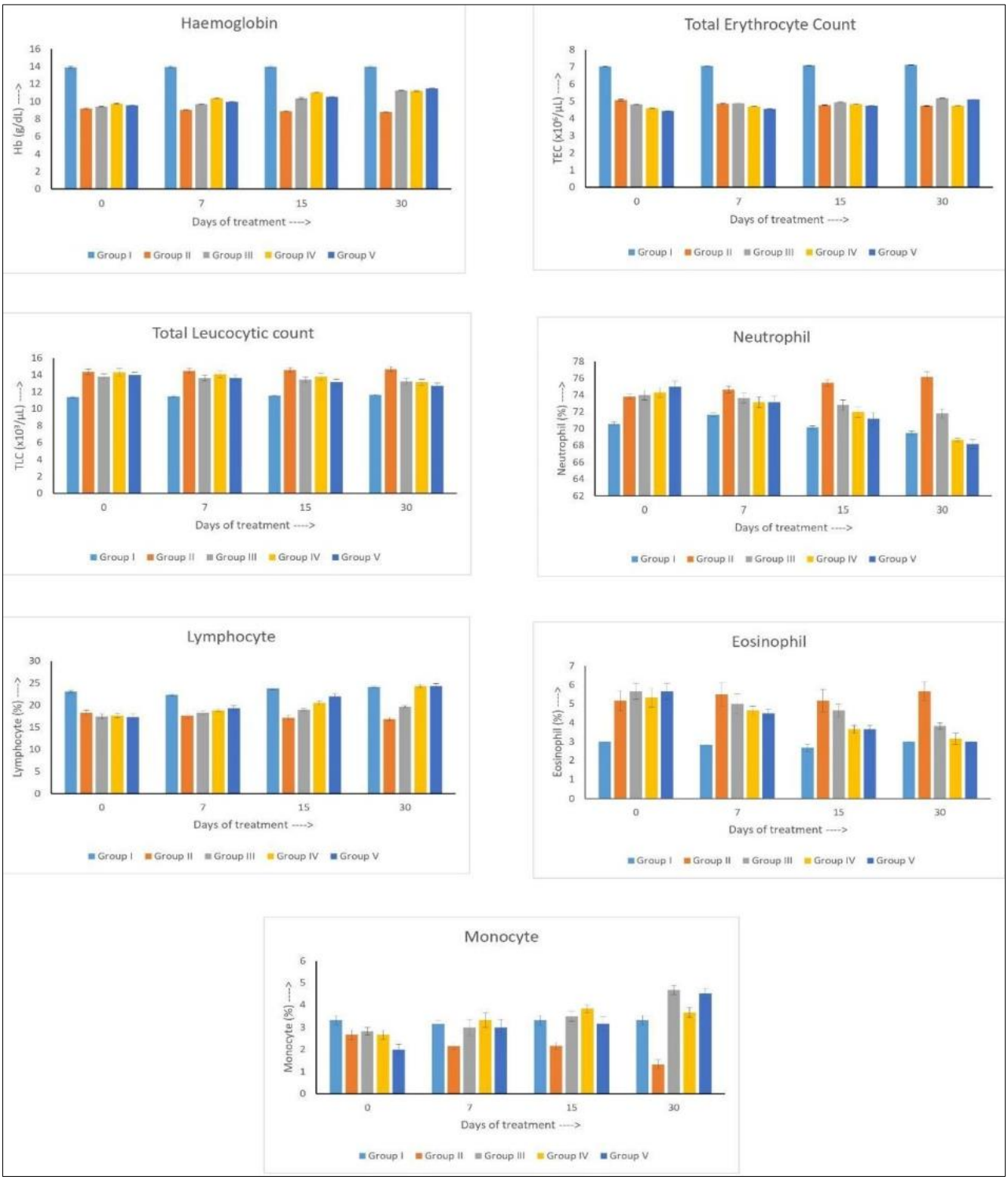

Fig 1: Haematological changes in control and treatment groups of pups

3.3 Serum biochemical parameters: The results of the haematological observations in different groups of pups have been presented in Table 3 and Figure 2. The table shows that there were significantly (p<0.01) lower levels of total serum protein on 0th day in Groups 2, 3, 4 & 5 in comparison to the healthy control puppies of Group 1. These findings are in conformity with Brar *et al.* (1993) [36] with toxocariosis and it might be due to impaired absorption of nutrients from the intestine and poor intake of food. In Group 2, there was further decrease of the values while in Groups 3, 4 & 5, the TSP values increased markedly at the end and best improvement was noted in Group 5.

The blood glucose values of the infected pups of Groups 2, 3, 4 & 5 was significantly (p<0.01) low on 0th day than the puppies of Groups 1 and agrees with the observations of Brar *et al.* (1993) [36] and it might be due to impaired absorption of nutrients in the intestine following *Toxocara spp*. infection. But with anthelmintic treatments in Groups 3, 4 & 5, the blood glucose levels increased significantly (p<0.01) at the end of the experiment when there was still further deterioration in Group 2.

The serum bilirubin levels in Groups 2, 3, 4 & 5 increased moderately on 0th day than the healthy pups of Group 1 and in agreement with the findings of Mondal *et al.* (2001) [4] who also noted mild increase of bilirubin levels in pups with toxocariosis and opined *Toxocara spp*. infection produces hepatic insufficiency and increases the serum bilirubin levels. However, following therapy with anthelmintics along, the values gradually improved in different treated groups. The mean values of AST in Groups 2, 3, 4 & 5 were significantly high (p<0.01) on 0th day in comparison to Group 1 and simulated with the observations of Mondal *et al.* (2001) [4] who also recorded higher AST values in piglets with *Toxocara spp*. infection and it might be due to hepatic insufficiency. But following treatment in Groups 3, 4 and 5, the AST values declined significantly (p<0.01) and became almost normal at the end of the experiment. Similarly, the ALT values were also found significantly high (p<0.01) on 0th day in the infected groups (Groups 2, 3, 4 and 5) and confirmed the findings of Mondal *et al.*, 2001 [4] in piglets with toxocariosis. However, in the treated groups (Groups 3, 4 & 5) the values almost became normal on 30th day, while it still remained significantly (p<0.01) elevated in Group 2.

Significantly lower levels of serum globulin were observed in Groups 2, 3, 4 and 5 on 0th day and it simulated with the findings of Rao *et al.* (1995) [16] who also noted decreased globulin levels in *Toxocara canis* infected pups and dogs, respectively. Following treatment in Groups 2, 3, 4 and 5, The mean albumin values were significantly lower (p<0.01) in all infected groups on 0th day and simulated with the observations of Rao *et al.* (1995) [16] who noted a decrease in serum albumin levels in dogs with *Toxocara canis* infection, which might be due to declination of diet intake, malabsorption and protein losing enteropathy owing to toxocariosis. However, following treatments in Groups 3, 4, and 5, the total serum albumin values became normal at the end of the experiment indicating control of *Toxocara* infection in these pups.

Table 3: Biochemical changes in control and treatment groups of pups

Sl. No.	Parameter	Group	Pre-treatment	Days of observation post-treatment		
			0th Day	7th Day	15th Day	30th Day
1	Total Serum Protein (g/dL)	I	6.96 ± 0.05^{a}	7.01 ± 0.03^{a}	6.98 ± 0.03^{a}	7.03 ± 0.02^{a}
		II	$4.90\pm0.09^{c}{}_{w}$	$4.90\pm0.09^{c}{}_{w}$	$4.58\pm0.07^{d}{}_{wx}$	$4.43\pm0.07^{d}{}_{x}$
		III	$5.20\pm0.05^{b}{}_{y}$	$5.33\pm0.05^{b}{}_{xy}$	$5.56\pm0.06^{c}{}_{wx}$	$5.76\pm0.06^{c}{}_{w}$
		IV	$4.95\pm0.04^{c}{}_{y}$	$5.26\pm0.04^{b}{}_{xy}$	$5.53\pm0.09^{c}{}_{x}$	$5.98\pm0.13^{bc}{}_{w}$
		V	$4.93\pm0.04^{c}{}_{y}$	$5.28\pm0.09^{b}{}_{x}$	$6.05\pm0.05^{b}{}_{w}$	$6.20\pm0.03^{b}{}_{w}$
2	Serum Albumin (g/dL)	I	3.63 ± 0.04^{a}	3.61 ± 0.06^{a}	3.50 ± 0.05^{a}	3.56 ± 0.05^{a}
		II	$2.28\pm0.06^{b}{}_{w}$	$2.16\pm0.05^{d}{}_{wx}$	$2.10\pm0.03^{d}{}_{x}$	$2.05\pm0.02^{d}{}_{x}$
		III	$2.35\pm0.03^{b}{}_{y}$	$2.45\pm0.03^{c}{}_{y}$	$2.73\pm0.04^{c}{}_{x}$	$2.95\pm0.05^{c}{}_{w}$
		IV	$2.38\pm0.04^{b}{}_{z}$	$2.63\pm0.033^{bc}{}_{y}$	$2.86\pm0.03^{c}{}_{x}$	$3.01\pm0.03^{c}{}_{w}$
		V	$2.35\pm0.08^{b}{}_{y}$	$2.78\pm0.06^{b}{}_{x}$	$3.15\pm0.04^{b}{}_{w}$	$3.28\pm0.03^{b}{}_{w}$
3	Serum Globulin (g/dL)	I	3.33 ± 0.03^{a}	3.40 ± 0.04^{a}	3.45 ± 0.04^{a}	3.46 ± 0.06^{a}
		II	2.61 ± 0.09^{c}	2.56 ± 0.06^{c}	2.48 ± 0.06^{c}	2.40 ± 0.07^{c}
		III	2.85 ± 0.03^{b}	2.88 ± 0.04^{b}	2.83 ± 0.04^{b}	2.81 ± 0.03^{b}
		IV	$2.56\pm0.04^{c}{}_{x}$	$2.63\pm0.02^{c}{}_{x}$	$2.66\pm0.08^{bc}{}_{x}$	$2.96\pm0.10^{b}{}_{w}$
		V	$2.58\pm0.04^{c}{}_{x}$	$2.50\pm0.05^{c}{}_{x}$	$2.90\pm0.03^{b}{}_{w}$	$2.91\pm0.03^{b}{}_{w}$
4	Albumin:Globulin ratio	I	1.08 ± 0.02^{a}	1.06 ± 0.03^{ab}	1.02 ± 0.02^{ab}	1.03 ± 0.03^{a}
		II	0.88 ± 0.04^{b}	0.84 ± 0.02^{c}	0.85 ± 0.03^{c}	0.85 ± 0.03^{b}
		III	$0.83\pm0.02^{b}{}_{y}$	$0.85\pm0.02^{c}{}_{y}$	$0.97\pm0.03^{b}{}_{x}$	$1.04\pm0.02^{a}{}_{w}$
		IV	$0.93\pm0.03^{b}{}_{y}$	$0.99\pm0.01^{b}{}_{wx}$	$1.08\pm0.03^{a}{}_{w}$	$1.02\pm0.03^{a}{}_{wx}$
		V	$0.91\pm0.04^{b}{}_{x}$	$1.11\pm0.03^{a}{}_{w}$	$1.08\pm0.02^{a}{}_{w}$	$1.13\pm0.02^{a}{}_{w}$
5	Blood Glucose(mg/dL)	I	105.00 ± 2.04^{a}	105.50 ± 1.78^{a}	105.66 ± 2.02^{a}	106.33 ± 2.01^{ab}
		II	$69.00\pm0.51^{b}{}_{w}$	$67.50\pm0.50^{d}{}_{wx}$	$65.83\pm0.70^{c}{}_{xy}$	$64.33\pm0.55^{c}{}_{y}$
		III	$69.00\pm0.36^{b}{}_{z}$	$74.66\pm0.61^{c}{}_{y}$	$87.00\pm1.34^{b}{}_{x}$	$102.50\pm1.62^{b}{}_{w}$
		IV	$67.33\pm0.88^{b}{}_{z}$	$76.83\pm1.16^{c}{}_{y}$	$85.50\pm0.99^{b}{}_{x}$	$104.50\pm1.66^{ab}{}_{w}$
		V	$67.50\pm1.38^{b}{}_{y}$	$84.33\pm1.38^{b}{}_{x}$	$105.66\pm1.76^{a}{}_{w}$	$109.33\pm0.98^{a}{}_{w}$
6	Serum Bilirubin(mg/dl)	I	0.38 ± 0.02	0.40 ± 0.00^{b}	0.38 ± 0.01^{b}	0.40 ± 0.00
		II	0.43 ± 0.03	0.45 ± 0.02^{ab}	0.45 ± 0.02^{ab}	0.46 ± 0.02
		III	0.43 ± 0.02	0.41 ± 0.01^{ab}	0.48 ± 0.02^{a}	0.46 ± 0.02
		IV	0.45 ± 0.02	0.43 ± 0.02^{ab}	0.42 ± 0.02^{ab}	0.46 ± 0.02
		V	$0.46\pm0.02_{w}$	$0.48\pm0.02^{a}{}_{w}$	$0.48\pm0.02^{a}{}_{w}$	$0.40\pm0.00_{x}$
7	AST (IU/L)	I	22.50 ± 0.67^{b}	23.00 ± 0.44^{c}	23.33 ± 0.61^{c}	22.83 ± 0.70^{c}
		II	42.66 ± 1.70^{a}	43.83 ± 1.64^{a}	45.16 ± 1.60^{a}	45.83 ± 1.57^{a}
		III	$41.83\pm2.67^{a}{}_{w}$	$36.33\pm1.83^{b}{}_{wx}$	$31.33\pm1.08^{b}{}_{xy}$	$26.83\pm0.60^{b}{}_{y}$

		IV	$43.50\pm1.78^{a}_{w}$	$36.50\pm1.20^{b}_{x}$	$32.50\pm1.23^{b}_{x}$	$26.66\pm0.80^{bc}_{y}$
		V	$47.00\pm0.89^{a}_{w}$	$36.33\pm1.42^{b}_{x}$	$30.66\pm1.58^{b}_{y}$	$24.33\pm0.76^{bc}_{z}$
8	ALT (IU/L)	I	31.33 ± 1.08^{b}	32.33 ± 0.84^{d}	32.83 ± 0.40^{c}	33.50 ± 0.42^{bc}
		II	66.16 ± 2.98^{a}	67.83 ± 2.91^{a}	68.33 ± 3.14^{a}	69.50 ± 3.04^{a}
		III	$66.33\pm1.20^{a}_{w}$	$58.50\pm1.54^{b}_{x}$	$50.66\pm1.11^{b}_{y}$	$39.33\pm1.96^{b}_{z}$
		IV	$63.50\pm0.99^{a}_{w}$	$55.16\pm0.79^{bc}_{x}$	$47.16\pm1.16^{b}_{y}$	$35.66\pm1.42^{bc}_{z}$
		V	$68.83\pm1.66^{a}_{w}$	$48.83\pm2.45^{c}_{x}$	$38.00\pm1.39^{c}_{y}$	$30.50\pm1.68^{c}_{z}$
9	Creatinine (mg/dL)	I	0.98 ± 0.07^{ab}	0.98 ± 0.09^{b}	0.98 ± 0.10^{bc}	1.05 ± 0.11^{b}
		II	$1.33\pm0.05^{b}_{x}$	$1.21\pm0.060^{ab}_{wx}$	$1.35\pm0.04^{a}_{w}$	$1.40\pm0.03^{a}_{w}$
		III	$1.45\pm0.04^{a}_{w}$	$1.31\pm0.04^{a}_{wx}$	$1.26\pm0.06^{a}_{wx}$	$1.18\pm0.06^{ab}_{wx}$
		IV	$1.41\pm0.05^{a}_{w}$	$1.33\pm0.04^{a}_{wx}$	$1.23\pm0.04^{ab}_{x}$	$1.05\pm0.04^{b}_{y}$
		V	$1.43\pm0.07^{a}_{w}$	$1.25\pm0.03^{a}_{wx}$	$0.93\pm0.03^{c}_{x}$	$0.90\pm0.05^{c}_{y}$

Mean bearing different superscripts (a, b, c, d and e) in a column differ significantly ($p<0.01$).
Mean bearing different subscripts (w, x, y and z) in a row differ significantly ($p<0.01$).

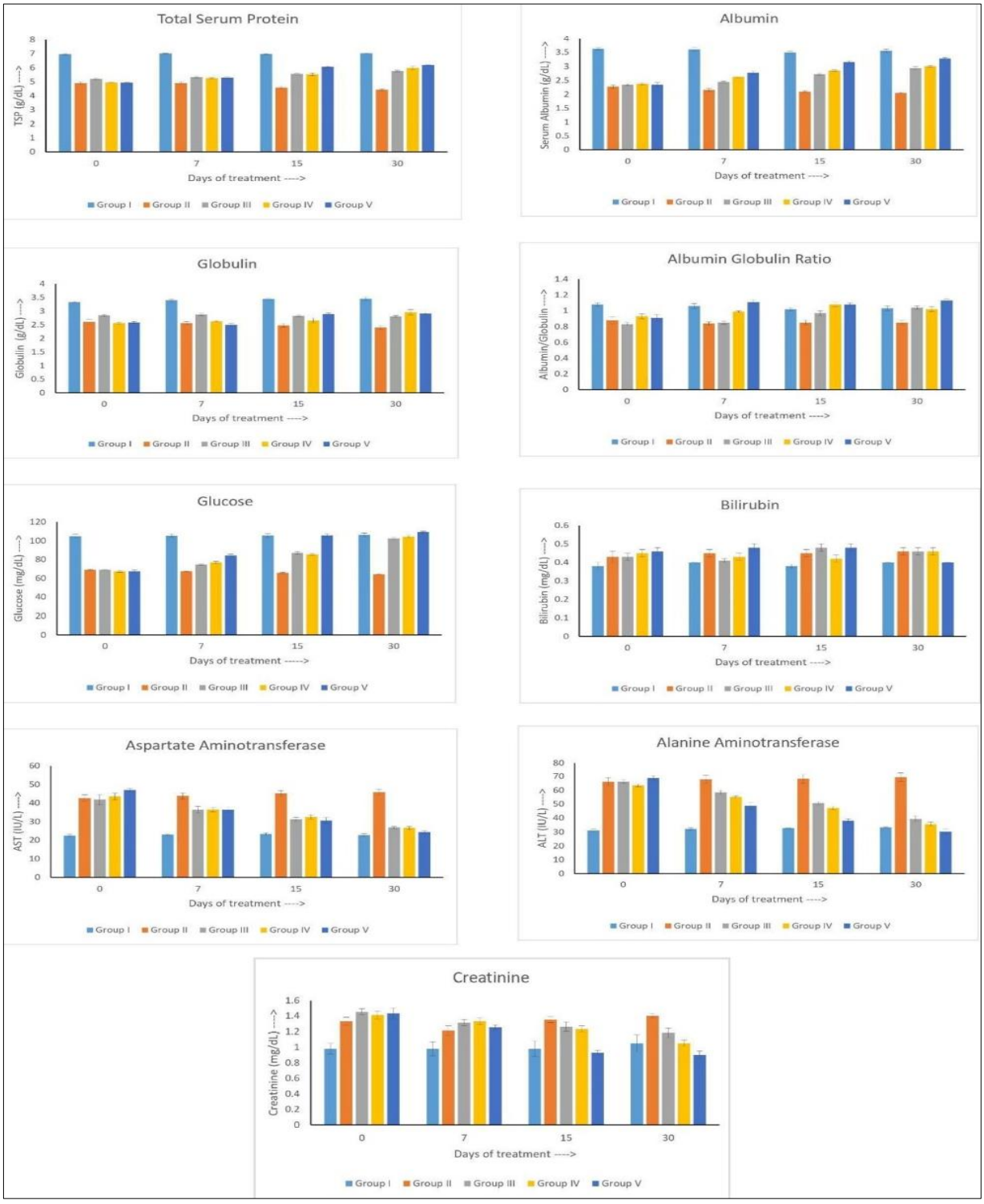

Fig 2: Biochemical changes in control and treatment groups of pups

The serum creatinine values of the healthy control pups of Group 1 varied from 0.98±0.07 mg/dL to 1.05±0.11 mg/dL during the entire experiment, and simulated with the observations of Chauhan *et al*. (2003) [37]. But in the infected pups, the value significantly increased (p<0.01), in a study of parasitic infected rats Shalaby *et al*. (2003) [38] noted elevated levels of serum creatinine in them and opined parasitisation induces the increase of creatinine levels in blood and also might be due to moderate to severe nephritis or toxic nephrosis.

3.4 Therapeutic trail and Efficacy

The eggs per gram of faeces and percent efficacy in different groups have been presented in Table 4. The table shows that the healthy puppies of Group 1 did not reveal any ova throughout the experiment while the infected puppies in Group 2 showed gradual increase of the same. But with different treatments in Groups 3, 4, and 5, the EPG values declined gradually and became nil at the end of the experiment.

Table 4: Eggs per gram and Percent Efficacy in pups before and after treatment

Group	Eggs Per Gram					Percent Efficacy (%)
	Pre-treatment	Days of observation post-treatment				
	0th Day	3rd Day	7th Day	15th Day	30th Day	
I	0	0	0	0	0	
II	$1200.00\pm57.73^{a}_{y}$	$1316.66\pm47.72^{a}_{xy}$	$1450.00\pm61.91^{a}_{wx}$	$1533.33\pm49.44^{a}_{wx}$	$1666.67\pm55.77^{a}_{w}$	
III	$1216.66\pm60.09^{a}_{w}$	$200.00\pm25.81^{b}_{x}$	$150.00\pm22.36^{b}_{xy}$	$33.33\pm21.08^{b}_{yz}$	$0\pm0^{b}_{z}$	89.49
IV	$1266.67\pm42.10^{a}_{w}$	$166.66\pm21.08^{b}_{x}$	$83.33\pm30.73^{b}_{xy}$	$16.66\pm16.66^{b}_{y}$	$0\pm0^{b}_{y}$	92.98
V	$1300.00\pm36.51^{a}_{w}$	$133.33\pm21.08^{b}_{x}$	$50.00\pm22.36^{b}_{xy}$	$0\pm0^{b}_{y}$	$0\pm0^{b}_{y}$	95.29

Mean bearing different superscripts (a, b, c, d and e) in a column differ significantly (p<0.01).
Mean bearing different subscripts (w, x, y and z) in a row differ significantly (p<0.01).

The table shows that, the overall efficacy of Pyrantal pamoate was 89.49%. Sharma *et al.* (2006) [30] reported 80-100% efficacy using a combination of Pyrantel pamoate, Fenbendazole and Praziquantel against toxocariosis. Group 4 puppies, which were treated with Albendazole showed overall percent efficacy of 92.98%. Similar efficacy ranging from 80-100% was reported by Tiwari (1994) [39] against *Toxocara canis* infection in dogs. Group 5 puppies, who were treated with Pyrantel embonate, Oxantel embonate and Praziquantel showed 95.29% efficacy. The Pyrantel and Oxantel are the depolarizing neuromuscular blocking agents in nematode parasites and produces paralysis in them. Praziquantel initiates rapid muscular contraction by altering the ionic balance of muscle cell and causes spastic paralysis of the worms and are eliminated. Sinha *et al.* (2009) [40] used a product containing Pyrantel pamoate, Fenbendazole and Praziquantel against nematodiasis in dogs and found effective. Considering the clinical improvements, haemato-biochemical changes and therapeutic efficacies, these drugs were found effective in treatment of *Toxocara spp*. infection in pups.

4. Conclusion

From the findings of the present study, the following conclusions were drawn: (i) The overall incidence of *Toxocara canis* infection in and around Kolkata city was 38.18%, (ii) the incidence rate was highest during the monsoon season (64.29%) and lowest in the winter season (9.52%), (iii) sex-wise prevalence rate was higher in the males (66.67%) than females (33.33%), (iv) incidence was higher (54.77%) in the age group of 1-3 months and lower (45.23%) in the age group of 4-6 months of age, (iv) non-descript (local) breeds of puppies (61.90%) were most commonly infected than the pure breed puppies, (v) a decrease of Hb, TEC, lymphocyte count, total serum protein, blood glucose, albumin, albumin: globulin ratio and increase in TLC, neutrophil, globulin, ALT, AST, total serum bilirubin, creatinine and eosinophil counts were noted in the pre-treatment infected groups of pups which changed in the post treatment observations, and (vi) all the three anthelmintics were found effective against the *Toxocara spp.* infection in the pups. However, for considering the best efficacy, the combination of Praziquantel, Pyrantel embonate and Oxantel embonate at a dose rate of 1 tab/10 kg b.wt. as single dose can suitably be recommended for treatment of toxocariosis in pups.

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