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Comparative efficacy of doxycycline and imidocarb diprionate in treatment of ehrlichiosis in dogs

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Abstract
Retrospective study evaluated the efficacy of Doxycycline and Imidocarb dipropionate in ehrlichia infected dogs. Based on history, clinical signs, peripheral blood smears examination, buffy coat smear examination, anigen Rapid *E.canis* Ab Test kit, and nested PCR, 17 dogs were diagnosed as canine ehrlichiosis among them Twelve dogs were randomly selected into 2 groups; Group II : Doxycycline and lactic acid bacillus (Doxypet) @10mg/kg b.wt. Per orally once in a day (n=6), Group III: Imidocarb dipropionate 12%w/v injection (Imicarb) @6.6 mg/kg b.wt, intramuscularly twice 14 days apart (n=6). The results revealed that magnitude of improvement and reversal of normalcy in certain clinical and various haemato biochemical parameters was far greater in Group II than Group III.

Keywords: Canine ehrlichiosis, Comparative efficacy, Doxycycline, Imidocarb diprionate

Introduction
Canine ehrlichiosis is mainly characterized by fever, anorexia, weakness, epistaxis, lymphadenopathy, tick infestation and ocular changes. Anemia, thrombocytopenia, hypoalbuminemia, hyper globulinemia, increased alkaline phosphatase (ALP) and alanine aminotransferase (ALT) are the major hemato-biochemical alterations. The disease is diagnosed by clinical signs, hematologic abnormalities, demonstration of morulae in peripheral monocytes, and detection of serum antibodies to *E. Canis* by the indirect immunofluorescence antibody (IFA) test [1]. Molecular techniques like PCR serve as a sensitive and specific diagnostic tool and it's highly reproducible and aids in fast interpretation of the disease. An early diagnosis of the disease is imperative to ensure successful treatment and good prognosis. The treatment of *E. canis* infection consists of antibacterial agents and supportive care. Efficacious drugs include tetracyclines (Doxycycline, Minocycline, Tetracycline and Oxytetracycline) and chloramphenicol. Anti protozoal drug Imidocarb dipropionate has been used in treating *E. canis* infections, this may be used in conjunction with tetracyclines or as alone. Generally, the earlier treatment of acutely infected dogs is initiated, the more favorable the prognosis and outcome. Dogs in the chronic phase are generally unresponsive to treatment because of the multi systemic disease changes and the severe myelo suppression [2].

Materials and Methods
The retrospective study was conducted on 750 dogs which was referred to small animal medicine ward, NTR College of Veterinary Science, Gannavaram and NTR Super Specialty Veterinary Hospital, Vijayawada, of them 17 dogs were diagnosed as canine ehrlichiosis by using different diagnostic techniques like peripheral blood smears examination, buffy coat smear examination, anigen Rapid *E.canis* Ab Test kit, and nested PCR. Twelve dogs which were positive for *E. canis* were randomly selected into 2 groups (Group II, Group III) each consisting of 6 dogs and data on clinical examination findings, haemato-biochemical parameters were registered before and after treatment in the both groups. A total 6 apparently healthy dogs were considered as health control (Group I). Group II were received Doxycycline and lactic acid bacillus (Doxypet, Savavet) @10mg/kg b.wt. Per orally once in a day, whereas group III were received Imidocarb dipropionate 12%w/v injection (Imicarb, Savavet). Additionally supportive therapy was given for both the groups (II and III) with hepatoprotactent (Ventriliv-pet, Venkys), Iron supplementation (Fe-folate, Venkys), Platelet enhancer (Plato grow, Ek-Tek Pharma) @8ml / day each orally. The owner way further advised to use Fipronil spot-on (Nay flee plus) and 1% cypermethrin shampoo (Clinar-M) to

control ticks. Comparative efficacy of doxycycline and imidocarb diprionate in treatment of ehrlichiosis in dogs based on clinical recovery, absence of morula stage in blood smear examination and hemato-biochemical changes were recorded and presented.

Results and Discussion

The results of the present study recorded and presented in Table 1, 2, 3 and 4.

The most frequently observed clinical findings in canine ehrlichiosis were anorexia and tick infestation history, lymphadenopathy, Pale conjunctival mucus membrane, splenomegaly and pyrexia, hemorrhagic tendencies like melena, petechial hemorrhages and epistaxis. Other clinical signs such as respiratory distress, corneal opacity, ataxia, recumbency, limb or scrotal edema and digestive disturbances like emesis were less frequently recorded (Table 1). The present observations corroborated with observation of [3]. Canine ehrlichiosis diagnosed by using different diagnostic techniques like peripheral blood smears examination, buffy coat smear examination, anigen Rapid *E.canis* Ab Test kit, and nested PCR.

In the present study clinical trial was conducted to study the efficacy of two drugs, doxycycline and Imidocarb propionate based on the magnitude of clinical recovery and improvement in haemato-biochemical profile.

The clinical recovery was found to be much faster in doxycycline Group, after initiation of therapy at each stage of observation, the magnitude of improvement in clinical parameters were within the period of 3-7 days as compared to 4-10 days in Imidocarb group. However, imidocarb treated dogs with acute stage of infection responded well as compared to the dogs with later stage of infection (Table 1 & 2). The results revealed that both the drugs were able to combat the canine ehrlichiosis however, critical evaluation of the response elicited by these drugs in treated canines indicated that doxycycline was found to be more efficacious than Imidocarb dipropionate. These findings were agreement with reports of Sainz *et al. (*2015) [4], who observed that doxycycline is an effective drug against ehrlichiosis in dogs with quick improvement. While non-agreement with reports of Price and Dolan (1980) [5], they reported superior findings in imidocarb group when compared to tetracycline hydrochloride.

Hematology revealed significant variations in the leukocyte indices might be due to collection of blood samples at different stage of infection [6]. In current study, lymphocytopenia is suggestive of myelo suppression, granylocytosis might be due to neutrophilia, *E.canis* cause immune suppression which originates secondary bacterial infection [7,8]. Anemic changes (decreased TEC, Hb, HCT) and thrombocytopenia were significant ($P<0.01$) when compared with healthy control as endorsed by earlier workers [9,10]. Decreased Hb and TEC could be due to epistaxis, petechial hemorrhages and bone marrow hypoplasia by the parasites leading to impaired production of cellular components of blood [11]. Thrombocytopenia occurs due to increased platelet consumption and decreased platelet half-life due to immune mediated splenic sequestration and destruction [12]. Post treatment haematological profile revealed that significantly ($P\leq0.01$) improved WBC, lymphocyte, erythrocyte, platelet counts whereas, monocyte, granulocyte, hemoglobin and HCT counts were significantly ($P\leq0.05$) reached towards normalcy in doxycycline treated group. While in imidocarb propionate treated Group WBC, lymphocyte, monocyte, erythrocyte, hemoglobin, HCT, platelet counts were significantly ($P\leq0.05$) towards normalcy and no statistical improvement in granulocyte. These results were agreed with Bhadesiya *et al*., (2015a) [13].

As compared to healthy control, the serum biochemistry revealed significantly ($P<0.01$) decreased levels of total serum protein and serum albumin and it was in line with previous reporters (Mylonakis *et al.*, 2010 and Kottadamane *et al.* 2016) [14, 15] where there was significant increase in the BUN, creatinine, ALT and ALP levels and these findings were comparable with the result of kasondra *et al.* (2016) [10]. However, there is no statistical difference in globulin level and this finding agreed with Mylonakis *et al.* (2010) [14]. In contrary, Heeb heather *et al.* (2003) [16] reported hyperglobulinemia. The hypoproteinemia, hypoalbuminemia may be due to anorexia, peripheral loss to edematous inflammatory fluid as consequence of vaculitis [17]. Decreased protein production because of concurrent liver diseases or proteinuria [6]. *E. canis* affecting other internal organs like liver and kidney, hence there was elevation in ALT, ALP, blood urea nitrogen and creatinine level. The elevation of ALT and creatinine levels might be due to immune complex-mediated glomerulonephritis indicating renal involvement in dogs with ehrlichiosis [18]. Post treatment biochemical profile revealed that Total serum protein, Serum Albumin, ALT, ALP, BUN, Creatinine were high significantly ($P\leq0.01$) and Serum globulin count significantly ($P\leq0.05$) towards normalcy in doxycycline treated group (Group-II). However, total protein, albumin, ALP, BUN, creatinine were significantly ($P\leq0.05$) and ALT was high significantly ($P\leq0.01$) changed towards normalcy in imidocarb treated Group (Group-III). This findings were supported with Kottadamane *et al.,* (2016) [15], those who reported superior results in clinico-hemato biochemical findings with doxycycline and supportive therapy. On 30th day both groups of laboratory recovery way also confirmed by examination buffy coat revealed no parasites.

Conclusion

Based on above results it is concluded that doxycycline is better choice for the treatment of canine ehrlichiosis than imidocarb dipropionate in the present research, equipotent in eliminating *E. canis* infection from blood on thirty days post treatment.

Plate 1: Clinical improvement in Group-II dogs

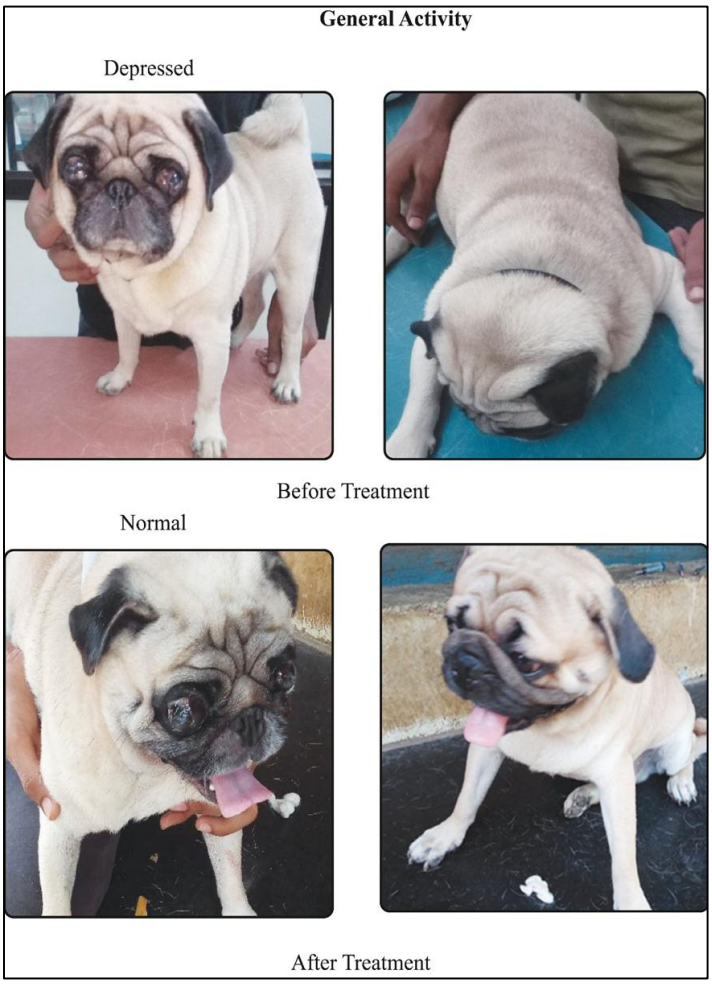

Plate 2: Clinical Improvement in Group –III dogs

Table 1: Clinical improvement in Group II

		BT	After treatment								
Parameter		**0th day**	**1st day**	**2nd day**	**3rd day**	**4th day**	**5th day**	**6th day**	**7th day**	**15th day**	**30th day**
General activity	Dullness	6	6	5	4	3	3	1	1	-	-
	Normal	-	-	1	2	3	3	5	5	6	6
Temperature	Pyrexia	5	5	2	2	-	-	-	-	-	-
	Normal	1	1	4	4	6	6	6	6	6	6
Appetite	Anorexia	6	6	4	4	1	1	1	1	1	1
	Normal appetite	-	-	-	-	5	5	6	6	6	6
Limb/scrotal edema	Present	1	1	1	1	1	1	1	1	-	-
	Improvement	-	-	-	-	-	-	-	-	1	1
Conjunctival mucous membrane	Pale	5	5	5	5	5	5	5	5	3	-
	Normal	1	1	1	1	1	1	1	1	3	6
Lymph nodes	Lymphadenopathy	6	6	6	6	6	6	5	4	-	-
	Normal	-	-	-	-	-	-	1	2	6	6
Hemorrhagic tendencies	Epistaxis	2	2	-	-	-	-	-	-	-	-
	Normal			2	2	-	-	-	-	2	2
	Petechial hemorrhage	3	3	3	3	3	3	3	3	-	-
	Normal	-	-	-	-	-	-	-	-	3	3
	Melina	4	4	4	3	2	-	-	-	-	-
	Normal	-	-	-	1	2	4	-	-	4	4
Emesis	Present	1	1	-	-	-	-	-	-	-	-
	Absent			1	1	1	1	1	1	1	1
Respiratory problems	Present	2	2	2	2	-	-	-	-	-	-
	Improvement	-	-	-	-	2	2	2	2	2	2
Corneal opacity	Present	2	2	2	2	2	2	2	2	-	-
	Improvement	-	-	-	-	-	-	-	-	2	2

Table 2: Pre and post therapeutic hemato-biochemical profile in Group II (Mean+S.E)

Parameter	Group -1(control group)	Group –II (n=6)	
		Before therapy 0th day	After therapy 30th day
** WBC (10^3/ µl)	9.98±0.73	22.48± 2.10	14.30± 0.85
** Lymphocyte (%)	17.27± 0.85	12.93± 0.71	17.9± 0.51
* Monocyte (%)	4.22± 0.32	5.90± 0.76	4.30± 0.34
* Granulocyte (%)	78.08± 0.59	81.17± 0.91	77.42± 0.81
** Erythrocyte (10^6/µl)	6.83± 0.24	3.93± 0.67	5.73± 0.48
* Hemoglobin (g/dl)	15.08± 0.67	8.30± 1.38	10.93± 1.30
* HCT (%)	40.72±5.93	27.28± 3.68	35.55± 2.65
** Platelet (10^3/ µl)	326.33±24.94	80.83± 23.05	288.17± 16.94
Total serum protein(g/dl)**	6.55± 0.22	4.73 ± 0.33	5.52± 0.42
Serum Albumin (g/dl)**	2.93± 0.10	0.91± 0.09	1.9± 0.14
* Serum Globulin (g/dl)	3.62± 0.19	3.82± 0.27	3.63± 0.33
** ALT (IU/L)	30.13±3.86	93.37± 9.53	31.40± 3.05
** ALP (IU/L)	32.41± 3.5	113.91± 1.43	43.87± 6.40
** BUN (mg/dl)	19.50± 1.06	32.58 ± 5.10	20.26± 1.7
** Creatinine (mg/dl)	0.88± 0.07	1.88± 0.23	1.0± 0.05

**- Statistically highly significant ($P \leq 0.01$)
*- Statistically significant ($P \leq 0.05$)

Table 3: Clinical improvement in Group III

		BT	After treatment							
Parameter		0th day	1st day	2nd day	3rd day	4th day	5th day	6th day	7th day	15th day	30thday
General activity	Dullness	6	6	6	6	6	5	4	4	-	-
	Normal	-	-	-	-	-	1	2	2	5	6
Temperature	Pyrexia	6	4	1	1	-	-	-	-	-	-
	Normal	-	2	5	5	6	6	6	6	6	6
Appetite	Anorexia	6	6	5	5	5	3	3	3	-	-
	Normal appetite	-	-	1	1	1	3	3	3	6	6
Conjunctival mucous membrane	Pale	6	6	6	6	6	6	6	6	4	-
	Normal	-	-	-	-	-	-	-	-	2	6
Lymph nodes	Lymphadenopathy	6	6	6	6	6	4	4	4	-	-
	Normal	-	-	-	-	-	2	2	2	6	6
Hemorrhagic tendencies	Epistaxis	3	3	2	-	-	-	-	-	-	-
	Normal	-	-	1	3	3	3	3	3	3	3
	Petechial hemorrhage	2	2	2	2	2	2	2	2	-	-
	Normal	-	-	-	-	-	-	-	-	2	2
	Melina	4	4	4	4	4	2	2	2	-	-
	Normal	-	-	-	-	-	2	2	2	4	4
Respiratory distress	Present	1	1	1	1	1	1	1	1	-	-
	Improvement	-	-	-	-	-	-	-	-	1	1

Table 4: Pre and post therapeutic hemato-biochemical profile in Group III (Mean+S. E)

Parameter	Group -1 (control group)	Group –III (n=6)	
		Before therapy (0th day)	After therapy (30th day)
* WBC (10^3/ µl)	9.98±0.73	20.52± 2.43	13.85±0.72
* Lymphocyte (%)	17.27± 0.85	14.18± 2.13	18.67± 0.75
* Monocyte (%)	4.22± 0.32	6.33± 0.87	4.28± 0.46
Ns Granulocyte (%)	78.08± 0.59	79.48± 1.59	77.05± 0.83
* Erythrocyte (10^6/µl)	6.83± 0.24	3.59±0.26	4.35± 0.52
* Hemoglobin (g/dl)	15.08± 0.67	7.37± 0.63	9.12±1.13
* HCT (%)	40.72±5.93	24.05±1.72	33.75± 4.11
* Platelet (10^3/ µl)	326.33±24.94	67.33± 18.51	202±11.85
* Total serum protein	6.55± 0.22	5.25±0.42	5.38±0.42
* Serum Albumin	2.93± 0.10	1.08±0.19	1.31±0.16
* Serum Globulin	3.62± 0.19	4.18±0.26	4.02±0.28
** ALT	30.13±3.86	94.80±9.86	69.90±9.56
* ALP	32.41± 3.5	120.14±6.15	70.87±15.84
* BUN	19.50± 1.06	29.72±3.78	20.08±2.68
* Creatinine	0.88± 0.07	1.91±0.15	1.27±0.98

References

1. Waner T, Strenger C, Keysary A. Comparison of a clinic-based ELISA test kit with the immunofluorescence test for the assay of *Ehrlichia canis* antibodies in dogs. Journal of Veterinary Diagnostic. Investigation. 2000;12(3):240-244.
2. Harrus S, Waner T, Avidar Y, Bogin, E, Peh HC, Bark H. Serum protein alterations in canine ehrlichiosis.

Veterinary Parasitology. 1996;66(3-4):241-249.
3. Waner TA, Keysary H, Bark Sharabani E, Harrus S. Canine monocytic ehrlichiosis – an overview. Journal of Israel Veterinary Medical Association. 1999;54:231-254.
4. Sainz A, Roura X, Miro G, Estrada-Pena A, Kohn B, Harrus S. Guideline for veterinary practitioners on canine ehrlichiosis and anaplasmosis in Europe. Parasites & Vectors. 2015;8(1):75.
5. Price JE, Dolan TT. A comparison of the efficacy of imidocarb dipropionate and tetracycline hydrochloride in the treatment of canine ehrlichiosis. The Veterinary Record. 1980;107(12):275-277.
6. Shimon Harrus, Trevor Waner, Mark Neer T. from Greene E infectious diseases of dog and cat fourth edition text book, Elesevier saunders. Ehrlichia and Anaplasma Infections. 2006;26:227-237.
7. Waner T. Hematopathological changes in dogs infected with *Ehrlichia canis.* Israel Journal of Veterinary Medicine. 2008;63(1):19.
8. Dixit AK, Dixit P, Shukla PC. Canine monocytic ehrlichiosis and its therapeutic management in a dog. Intas Polivet. 2012;13(1):140-1.
9. Devi S, Saxena A, Singh RD, Jadhav KM. Clinical management of canine monocytic ehrlichiosis (CME) and associated epistaxis-a study in 6 patients. Intas Polivet 2015;1;16(2).
10. Kasondra Arjun, Snehil Gupta, Gamit Amit Bhai Bharat Bhai, Vijesh Kumar Saini. Therapeutic management of canine ehrlichiosis with aid of blood transfusion: a case report. Journal of Parasitic Diseases. 2016;41(2):395-397.
11. Neer TM, Breitschwerdt EB, Greene RT, Lappin MR. Consensus statement on ehrlichial disease of small animals from the infectious disease study group of the ACVIM. Journal of Veterinary Internal Medicine. 2002;16(3):309-15.
12. Sangeetha SG, Ajith Y, Dixit SK, Reena KK. PCR Based Diagnosis and Clinical Management of Ehrlichiosis in a Dog. Intas Polivet. 2017;18(1):187-192.
13. Bhadesiya CM, Raval SK. Hemato-biochemical changes in ehrlichiosis in dogs of Anand region, Gujarat. Veterinary world. 2015a;8(6):713.
14. Mylonakis ME, KritsepiKonstantinou M, Dumler JS, Diniz PP, Day MJ, Siarkou VI, et al. Severe hepatitis associated with acute *Ehrlichia canis* infection in a dog. Journal of Veterinary Internal Medicine. 2010;24(3):633-8.
15. Kottadamane MRL, Dhaliwal PS, Singla LD. Diagnosis and treatment of canine monocytic ehrlichiosis in a boxer breed of dog-a case report. International Journal of Science, Environment and Technology. 2016;5(5):3099-3105.
16. Heeb heather L, Wilkerson MJ, Chun R, Ganta RR. Large granular lymphocytosis lymphocyte subset inversion, thrombocytopenia, dysproteinemia, and positive Ehrlichia serology in a dog. Journal of the American Animal Hospital Association. 2003;39(4):379-384.
17. Woody BJ, Hoskins JD. Ehrlichial Diseases of Dogs Veterinary Clinics of North America: Small Animal Practice. 1991;21(1):130-135.
18. Bhadesiya CM, Raval SK. Therapeutic efficacy of oxytetrycline, doxycycline and enrofloxacin against ehrlichiosis-a clinical study of 18 dogs. Intas Polivet. 2015b;16(2):345-350.