

Haemato-Biochemical Changes and Serum C-Reactive Protein Concentration in Dogs with Acute Renal Failure

S.Yogeshpriya*, Usha Narayana Pillai, S.Ajithkumar and N. Madhavan Unny

Department of Clinical Veterinary Medicine, College of Veterinary and Animal Sciences, Kerala Veterinary and Animal Sciences, Mannuthy, Thrissur – 680651, Kerala, India



Abstract

The present study was designed to evaluate the C - reactive protein and it's correlation with leptospirosis induced acute renal failure. A total of 52 sera samples collected from dogs showing clinical symptoms of acute renal failure were selected and screened for the leptospira antibodies by microscopic agglutination test (MAT). Out of which 36 positive cases of leptospirosis used for the present study. Physiological parameters were studied in all 52 patients. Blood and serum samples collected for further evaluation. Serum C-reactive protein (CRP) was estimated on first day using the turbidimetric assay by standard kits. Out of 52 cases of acute renal failure, 69.23% cases were confirmed as leptospirosis. General physical examination of dogs showed mean temperature of 103.01 ± 1.15 ($^{\circ}\text{F}$). Abdominal pain was observed in 24 out of 36 dogs. The mean heart rates (beats/ min) and respiratory rates (beats/ min) on the day of admission were 120 ± 12 and 25 ± 13 respectively. Markedly increased ESR values were observed. Correlations were obtained between CRP values and albumin, total leucocyte count, neutrophil, heart rate, respiratory rates and temperature by using Pearson correlation. P value of < 0.05 was considered statistically significant. From a clinical perspective, acute phase protein measurement is a useful clinical tool in veterinary medicine but further studies are required to assure their value in particular diseases, because the single acute phase protein, CRP should not be enough indicator for screening of leptospirosis in dogs.

Key words: C-Reactive protein, Leptospirosis, Pearson correlation

Introduction

Canine leptospirosis is a contagious disease of varying severity, from latent to acute, caused by spirochetes of the genus leptospira spp. This disease is biphasic, in

earlier is leptospiremia and followed by leptospiruria.

As a part of the innate host defense system the acute phase response (APR) occurs following a variety of nonspecific insults including infection, inflammation, immune-mediated disorders, neoplasia, and trauma (Ceron *et al.*, 2005). The APR includes changes in the concentrations of acute phase proteins (APPs) which includes positive acute phase proteins included C- reactive protein (CRP), haptoglobin (Hp), ceruloplasmin (Cp), serum amyloid A (SAA), -1 acid glycoprotein and fibrinogen, these are elevated mainly due to increased production in the liver (Nakamura *et al.*, 2008). Whereas albumin and transferrin are negative APPs. In the last decade, emphasis has been laid on application of blood tests for acute phase reactants to monitor human health as well as animal health in general affecting from specific diseases.

The objective of the current study was to measure serum CRP concentrations in dogs diagnosed with acute renal failure due to leptospirosis at presentation. Data were collected and then evaluated to determine if there was an association between CRP concentration with white blood cell (WBC) counts, temperature, albumin, neutrophils at presentation.

Materials and methods

In the present prospective study, all dogs had to have an acute onset of one or more of the following clinical signs to be included in the study such as depression, lethargy, weakness, anorexia, vomiting, diarrhea, and change in urine production. Serum samples were taken from 52 dogs presented with the clinical signs of acute renal failure at the University Veterinary Hospital, Mannuthy and Thrissur, Kerala from June 2011 to December 2011.

* Email: dryogeshpriya@gmail.com

Out of which 36 animals found to be positive for leptospirosis in Microscopic agglutination test (MAT). Relevant clinical materials were collected at the time of admission. Five ml of whole blood was collected from saphenous or cephalic vein of the selected dogs in dry glass vials containing EDTA @ 1-2 mg per ml as anticoagulants (Benjamin, 1985). About 10 ml of blood was collected in another vial on the day of admission to separate serum for biochemical examination. Sera thus separated were stored at -20°C till further analysis. Total erythrocyte count, hemoglobin, volume of packed red cells, total leukocyte count, differential leukocyte count and platelet count were estimated as per the method described by Schalm *et al.* (1975). Blood urea nitrogen, serum creatinine, serum total proteins, serum albumin, serum globulin and A: G ratio and serum C-reactive protein were estimated on first day using semiautomatic analyzer, as per the manufacturer's instructions and using standard kits. Data were analysed wherever indicated as per Snedecor and Cochran (1994). Correlations between concentration of CRP with white blood cell (WBC) counts, temperature, albumin, erythrocyte sedimentation rate at presentation were determined by Pearson correlation.

Results and discussion

A total of 52 dogs with the presumptive diagnosis of acute renal failure were initially enrolled in the study. In 36 dogs, the diagnosis of leptospirosis was confirmed through MAT and the data collection was considered adequate. These 36 dogs were included in the study as the diseased group. Additionally 12 healthy dogs were included in this study as a control. Leptospirosis is one of the most commonly occurring diseases of acute renal failure. CRP concentration in the clinically normal dogs ranged from 0-9 µg/ml (Galezowski *et al.*, 2010). There was significant difference in BUN, creatinine, ALT, total bilirubin between control and clinically affected dogs. Similar results obtained by Sherding (2000). Increased liver enzymes and hypoalbuminemia were observed (Table-1) with high levels of serum C-reactive protein as reported by Oliveira *et al.* (2010). Out of the 36 dogs, 36.8% had high liver enzymes, 27.8% had azotemia, 67.1% had leukocytosis and 59.7% had hypoalbuminemia.

Table: 1 Serum biochemistry in dogs affected with leptospirosis

Parameters	Healthy dogs	Clinical cases
Total protein (g/dl)	7.58 ± 0.03	7.18 ± 1.02
Albumin (g/dl)	4.26± 0.02	2.42 ± 0.86
Globulin (g/dl)	3.32± 0.01	4.52 ± 1.46
Creatinine (mg/dl)	0.68± 0.02	7.19 ± 6.96
Total bilirubin(mg/dl)	0.28± 0.06	1.475 ± 2.98
Direct bilirubin (mg/dl)	0.07± 0.03	1.01 ± 0.26
SGPT (U/L)	18.69± 2.4	91.90 ± 59.36
ALP (U/L)	21.34± 2.5	113.88 ± 96.25
C-Reactive protein (mg/l)	6.09 ± 1.9	30.18 ± 8.51

There was significant strong correlation was observed between CRP concentration and temperature of the dogs with leptospirosis ($r = 0.855$, $P < 0.01$).

Table 2: Haematological changes in dogs with Leptospirosis

Parameters	Group	
	Control	Clinical cases
Total Leukocyte count (X10 ³ /µl)	13.9 ± 0.56	23.2 ± 11.33
Neutrophils (%)	10.7 ± 0.56	79± 12.17
Lymphocytes (%)	3.03 ± 0.12	19.33± 12.23
Eosinophils (%)	0.14 ± 0.01	1.33± 1.16
Erythrocyte Sedimentation Rate (mm/hour)	3.00 ± 0.37	34.73± 26.09
Hemoglobin (gms %)	14.03 ± 0.80	12.89± 3.36
Total erythrocyte count (million/cumm)	7.02 ± 0.46	6.2± 1.89
Volume of packed cells (%)	38.13 ± 2.89	39.38± 10.53
Platelet counts (X10 ⁶ /µl)	3.73± 3.95	2.69± 1.10
Mean corpuscular volume (cubic meters)	71.01 ± 9.28	64.51± 12.54
Mean Corpuscular Haemoglobin (pg)	23.46 ± 3.22	20.09± 4.06
Mean Corpuscular Haemoglobin Concentration (%)	33.01 ± 2.07	32.2± 1.79

No correlation was observed between CRP concentration and albumin in dogs with leptospirosis. C-reactive protein concentrations were increased with low serum albumin concentrations in many dogs (Table-3). The CRP concentration was dissociated from neutrophils in many cases, and by using Pearson correlation only a weak correlation was observed ($r = 0.37$). Comparison of CRP concentration of the animals with high TLC to that of the normal TLC, the CRP concentration was higher in animals with high TLC (Table-2). Galezowski *et al.* (2010) concluded that the acute phase proteins such as C-reactive protein (CRP) had a higher diagnostic sensitivity than leucocyte count in detecting inflammation. Eckersall and Bell (2010); Sheahan *et al.* (2010) and Tvarijonaviciute *et al.* (2011) opined that the acute phase proteins (APPs) constituted a group of proteins whose blood concentration changed rapidly in response to stress, infection and tissue damage.

Table 3 Pearson correlation for CRP

Parameters	Mean±SD	Correlation	Results
TLC(/ mm ³)	23.2±11.33	0.94**	Strong
N (%)	79 ± 12.17	0.37*	Weak
Albumin (g/dl)	2.42± 0.86	-0.16	Negative
Temperature (°F)	103.01 ± 1.15	0.855**	Strong

* Correlation is significant at the 0.05 level (2-tailed).

** Correlation is significant at the 0.01 level (2-tailed).

Summary

C-reactive protein had the characteristic response of a sensitive APP and received most attention in canine medicine. However, the results showed that C-reactive protein was not useful to predict leptospiremia in the naturally infected dogs of this study; and although association between urinary C-reactive protein and seropositivity was observed, it should not be used as a unique test for leptospirosis. In conclusion, although C-reactive protein may be used as part of a screening profile, it should not be considered as indicator alone of leptospirosis screening in dogs.

Acknowledgement

The authors are thankful to the Dean, College of Veterinary and Animal Sciences, Mannuthy and Department of Clinical Veterinary Medicine for providing necessary facilities to carry out the work.

References

- Benjamin, M.M.1985.Outline of veterinary clinical pathology. Third edition. Indian reprint. *Kalyani Publishers, New Delhi*. pp.18-27.
- Ceron, J.J., Eckersall, P.D. and Subiella, S.M.2005. Acute phase proteins in dogs and cats: current knowledge and future perspectives. *Vet. Clin. Pathol.* **34**: 85-99.
- Galezowski, A.M., Snead, E.C.R., Kidney, B.A. and Jackson, M.L.2010. C-reactive protein as a prognostic indicator in dogs with acute abdomen syndrome. *J. Vet. Diagn. Invest.* **22**: 395-401.
- Nakamura, M., Takahashi, M., Ohno, K., Koshino, A., Nakashima, K., Setoguchi, A., Fujino, Y. and Tsujimoto, H.2008. C-reactive protein concentration in dogs with various diseases. *J. Vet. Med. Sci.* **70**: 127-131.
- Oliveira, S.T., Messick, J.B., Biondo, A.W., Santos, A.P., Guimaraes, A.M.S., Mohamed, A.S., Pires Neto, J.A.S., Dalmolin, M.L. and Gonzalez, F.H.D.(2010). Serum and urinary C-Reactive protein concentration in dogs with leptospirosis. *Acta Scientiae Veterinariae.* **38(3)**: 245-249.
- Schalm, O.W., Jain, N.C. and Corel, E.J. 1975. Veterinary haematology. 3rd edn. *Lea and Febiger, Philadelphia*. p.647.
- Sherding, R.G.2000. Leptospirosis, Brucellosis & other bacterial infectious diseases. In: Saunderson's Manual of Small animal Practice. *Birchard, S.J. and Sherding, R.G. W.B.Saunders Co., Philadelphia*, 128-132.
- Snedecor, G.W. and Cochran, W.G. (1994). Statistical Methods. 8th edn. *Iowa state University Press, Ames, Iowa*