RESEARCH ARTICLE

Haemato-Biochemical Studies Before and After Ovario-Hysterectomy in Bitches Affected with Pyometra

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Abstract

This study was conducted on 18 bitches; 10 affected with pyometra and 8 normal healthy bitches to evaluate and compare the haemato-biochemical profiles before and after surgical intervention. The blood samples were collected from confirmed cases of canine pyometra and healthy bitches just before and on day 8th and 15th after surgery. The mean values of haemoglobin, packed cell volume, total erythrocyte count, total platelet count were significantly ($p < 0.01$) lower, and erythrocyte sedimentation rate and total leucocyte count were significantly ($p < 0.01$) higher in bitches affected with pyometra as compared to healthy ones on day 0. These differences gradually narrowed down on day 8th and 15th post-operative with improvement in profile of operated pyometric bitches. The mean neutrophil count was significantly increased, whereas the values of lymphocytes, monocytes and eosinophils were decreased in bitches with pyometra than in healthy ones, which gradually normalized in post-operative period. Significantly ($p < 0.05$) higher values of plasma urea nitrogen (46.13 ± 6.79 vs. 24.32 ± 5.16 mg/dl), creatinine (1.92 ± 0.42 vs 1.14 ± 011 mg/dl) plasma total protein and globulin (4.87 ± 0.27 vs 2.85 ± 0.19 g/dl) with lower A:G ratio (0.51 ± 0.03 vs 0.99 ± 0.10) were recorded in bitches with pyometra as compared to healthy ones, which declined non-significantly by 8th and 15th day post-operative in affected bitches. There was significant decline in the plasma levels of total cholesterol and enzymes aspartate amino transferase and alanine aminotransferase in bitches with pyometra at 8th and 15th day post-operative as compared to pre-operative values, however the differences between healthy and affected bitches were statistically non-significant, perhaps due to wide variation in the profiles of individual animals. It is concluded that haemato-biochemical profile in conjunction with clinical signs can be used to predict the severity of canine pyometra.

Keywords: Bitch, Haematology, Neutering, Ovario-hysterectomy, Plasma Biochemistry, Pyometra.

Ind J Vet Sci and Biotech (2020): 10.21887/ijvsbt.16.(2,3,&4).10

Introduction

Pyometra is the most common and serious reproductive disorder that affects nearly one fourth of all adult intact female dogs during diestrus period (Johnston *et al*., 2001; Kida *et al*., 2006). Pyometra may be acute or chronic and is characterized by genital and systemic illness resulting in impaired homeostasis, cystic endometrial hyperplasia, infiltration of inflammatory cells and accumulation of uterine exudates (Kempisty *et al*., 2013). The pathogen most often isolated from uteri of bitches was *Escherichia coli* (Hagman, 2018). The reduced levels of Hb, PCV, TEC and platelet counts along with elevated ESR, TLC and polymorphonuclear (PMN) cells indicate toxaemia, whereas raised levels of leucocytes, PMN cells and declining lymphocytes point out recovery from toxemia (Gupta and Dhami, 2013). The safest and most effective treatment opined is ovariohysterectomy (Kumar and Saxena, 2018). The present study was carried out with the objective to evaluate and compare haemato-biochemical profiles, before and after surgical intervention of canine pyometra.

Materials and Methods

The study was carried out during the year 2019-20 at the College of Veterinary Science, AAU, Anand. The study covered total 18 bitches, irrespective of breed and age, with a history of pyometra (n = 10) and normal healthy bitches (n = 8). The condition was confirmed based on history, clinical signs. abdominal palpation, USG, radiography and laboratory findings, and the prognosis for surgical intervention was predicted.

For haematology and biochemical parameters, about 5-6 ml of blood was collected from cephalic vein in a vial containing EDTA. A few ml of blood was utilized for haematology on auto-blood analyzer (Abacus Junior Vets 5) and erythrocyte sedimentation rate (ESR) was determined by

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How to cite this article: Hadiya, H.D., Patel, D.M., Parikh, P.V., & Rao, N. (2020). Haemato-Biochemical Studies Before and After Ovario-Hysterectomy in Bitches Affected with Pyometra. Ind J Vet Sci and Biotech, 16(2,3,&4): 45-49.

Source of support: Nil

Conflict of interest: None.

Submitted: 26/07/2020 **Accepted:** 10/10/2020 **Published:** 25/12/2020

Westergreen method. The remaining blood was centrifuged at 3000 rpm for 15 minutes and plasma was stored at -20°C with a drop of sodium merthiolate (0.1%) until analyzed for various biochemical parameters, *viz.,* plasma urea nitrogen (PUN), plasma creatinine, total protein, albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST) and total cholesterol by employing assay kits and standard procedures on an auto-analyzer (CKK). The level of plasma globulin was obtained by subtracting albumin from total protein and A:G ratio was worked out for each sample.

The surgical intervention performed as per standard techniques in practice through a mid-line approach included ovario-hysterectomy in cases of pyometra and spaying/ neutering in normal healthy bitches. The data generated on haemato-biochemical profile were analysed using completely randomized design and 't' test to know the variation between groups, and between pre- and post-operative stages in bitches (Snedecor and Cochran, 1994).

Results and Discussion

The haemato-biochemical profiles recorded before surgical operation, and on 8th day and 15th day post-ovario-hysterectomy in bitches affected with pyometra and in normal healthy bitches underwent spaying are presented in Table 1-3.

Haematological Alterations

The mean values of haemoglobin (9.83 ± 0.96 vs. 13.65 ± 0.64 g/dl), packed cell volume (23.61 ± 1.74 vs. 40.53 ± 1.75 %) and total erythrocyte count (5.91 ± 0.44 vs. 8.17 ± 0.44 million/ cmm) were significantly ($p < 0.01$) lower in bitches with pyometra as compared to healthy ones on day 0, *i.e.,* pre-operatively and the differences gradually narrowed down on 8th and 15th post-operative day with improvement in values of ovario-hysterectomized bitches. The mean ESR in bitches with pyometra was significantly higher than normal healthy bitches presented for spaying (6.70 ± 0.48 vs. 3.13 ± 0.43 mm/hr) and the values did not come down to normal even after 15 days of operation in diseased bitches (Table 1).

The mean value of total platelet count (TPC) in bitches with pyometra was significantly lower than in healthy bitches on day 0 (167.60 ± 17.31 vs. 295.38 ± 13.94 thousand/cmm), which gradually and significantly improved by day 8th and 15th post-operative, yet it did not come to normal values of healthy status. The value of total leucocyte count (TLC) (36.38 ± 6.72 vs. 21.11 ± 3.61 thousand/ cmm) was significantly higher in bitches with pyometra as compared to normal healthy bitches particularly at day 0, which reduced gradually at day 8th and 15th post-operatively, but the differences post-operative with healthy ones were statistically non-significant. The Hb and TPC increased significantly at 8th/15th day post-operatively (Table 1). The haematological findings of the

Table 1: Haematological values before surgical intervention, and on 8th and 15th day post-operative in bitches with pyometra and normal healthy bitches

Blood parameters	*Day of operation*	*Pyometra /ovario-hysterectomy (n=10)*	*Normal healthy spaying (n=8)*	*'t' test*
Haemoglobin (Hb, g%)	0	9.83 ± 0.96^{b}	13.65 ± 0.64	**
	8	11.78 ± 0.86^{ab}	14.05 ± 0.32	*
	15	12.99 ± 0.72^{a}	14.65 ± 0.18	*
Packed cell volume (PCV, %)	0	23.61 ± 1.74	40.53 ± 1.75	**
	8	28.78 ± 2.06	40.25 ± 1.36	**
	15	30.57 ± 2.17	40.66 ± 1.19	**
Total erythrocyte count (TEC, ×10^{6}/cmm)	0	5.91 ± 0.44	8.17 ± 0.44	**
	8	6.69 ± 0.39	7.98 ± 0.29	*
	15	7.10 ± 0.32	7.80 ± 0.23	NS
Erythrocyte sedimentation rate (ESR, mm/hr)	0	6.70 ± 0.48	3.13 ± 0.43	**
	8	7.18 ± 0.48	3.38 ± 0.51	**
	15	7.62 ± 0.45	3.38 ± 0.29	**
Total platelet count (TPC, ×10^{3}/ cmm)	0	167.60 ± 17.31^{b}	295.38 ± 13.94	**
	8	222.90 ± 12.86^{a}	306.88 ± 14.33	**
	15	250.90 ± 11.65^{a}	330.13 ± 8.59	**
Total leucocyte count (TLC, ×10^{3}/ cmm)	0	36.38 ± 6.72	21.11 ± 3.61	*
	8	27.73 ± 4.23	21.55 ± 2.83	NS
	15	20.17 ± 2.09	17.34 ± 1.60	NS

Day 0 = Pre-operative, 8 = 8th day post-operative, 15 = 15th day post-operative.
** = highly-significant ($p < 0.01$), * = significant ($p < 0.05$), NS = non-significant ($p \geq 0.05$).
Means with different superscripts within column differ significantly ($p < 0.05$) for a trait.

Table 2: Differential leucocyte count before surgical intervention, and on 8th and 15th day post-operative in bitches with pyometra and normal healthy bitches

Differential leukocyte count (%)	*Day of operation*	*Pyometra /ovario-hysterectomy (n=10)*	*Normal healthy spaying (n=8)*	*'t' test*
Neutrophils (%)	0	76.50 ± 2.66[a]	64.25 ± 1.90	**
	8	67.70 ± 1.63[b]	62.13 ± 2.11	NS
	15	63.40 ± 0.87[b]	58.63 ± 1.50	*
Lymphocytes (%)	0	20.70 ± 2.49[b]	29.63 ± 2.38	*
	8	29.20 ± 1.55[a]	32.75 ± 2.56	NS
	15	33.50 ± 0.92[a]	36.75 ± 1.78	NS
Monocytes (%)	0	1.40 ± 0.34	3.09 ± 0.67	NS
	8	1.70 ± 0.30	3.00 ± 0.38	*
	15	1.60 ± 0.43	2.13 ± 0.36	NS
Eosinophils (%)	0	1.20 ± 0.13	2.79 ± 0.36	**
	8	1.20 ± 0.25	2.25 ± 0.28	*
	15	1.20 ± 0.20	2.31 ± 0.37	*
Basophils (%)	0	0.20 ± 0.13	0.25 ± 0.15	NS
	8	0.20 ± 0.13	0.13 ± 0.11	NS
	15	0.30 ± 0.15	0.19 ± 0.12	NS

Day 0 = Pre-operative, 8 = 8th day post-operative, 15 = 15th day post-operative.
** = highly significant ($p < 0.01$), * = significant ($p < 0.05$), NS = non-significant ($p \geq 0.05$).

Table 3: Plasma biochemical summary before surgical intervention, and on 8th and 15th day post-operative in bitches with pyometra and normal healthy bitches

Plasma Biochemistry	*Day of operation*	*Pyometra /ovario-hysterectomy (n=10)*	*Normal healthy spaying (n=8)*	*'t' test*
Plasma urea nitrogen (mg/dl)	0	46.13 ± 6.79	24.32 ± 5.16	*
	8	33.60 ± 6.18	19.30 ± 2.35	NS
	15	28.35 ± 5.18	15.61 ± 1.00	*
Plasma creatinine (mg/dl)	0	1.92 ± 0.42	1.14 ± 0.11	NS
	8	1.37 ± 0.25	0.90 ± 0.06	NS
	15	1.23 ± 0.28	0.93 ± 0.06	NS
Plasma total protein (g/dl)	0	7.30 ± 0.26	5.56 ± 0.29	**
	8	7.14 ± 0.16	6.44 ± 0.28	NS
	15	7.34 ± 0.13	6.64 ± 0.20	*
Albumin (g/dl)	0	2.44 ± 0.06	2.71 ± 0.19	NS
	8	2.79 ± 0.18	2.90 ± 0.07	NS
	15	2.88 ± 0.18	3.31 ± 0.17	NS
Globulin (g/dl)	0	4.87 ± 0.27	2.85 ± 0.19	**
	8	4.35 ± 0.16	3.55 ± 0.23	*
	15	4.46 ± 0.14	3.55 ± 0.20	**
A: G Ratio	0	0.51 ± 0.03	0.99 ± 0.10	**
	8	0.65 ± 0.06	0.84 ± 0.04	*
	15	0.66 ± 0.06	0.92 ± 0.11	NS
AST (IU/L)	0	55.16 ± 4.84[a]	41.07 ± 1.28[a]	*
	8	44.16 ± 4.63[ab]	33.55 ± 2.53[b]	NS
	15	36.64 ± 5.15[b]	25.81 ± 1.94[c]	NS
ALT (IU/L)	0	56.29 ± 12.35[a]	31.08 ± 2.72	NS
	8	35.96 ± 4.09[ab]	26.46 ± 1.92	NS
	15	28.92 ± 2.19[b]	21.61 ± 2.52	NS
Plasma total cholesterol	0	249.70 ± 8.50[a]	196.63 ± 21.92	NS
	8	226.30 ± 7.68[b]	230.88 ± 20.62	NS
	15	218.40 ± 7.37[b]	251.75 ± 16.81	NS

Day 0 = Pre-operative, 8 = 8th day post-operative, 15 = 15th day post-operative.
AST= Aspartate amino-transferase, ALT= Alanine amino-transferase.
** = highly significant ($p < 0.01$), * = significant ($p < 0.05$), NS = non-significant ($p \geq 0.05$

present study compared well with those of Dabhi *et al*. (2009), Baithalu *et al*. (2010), Lika *et al*. (2011), Gupta and Dhami (2013), Patil *et al.* (2013), Mohan *et al*. (2015) and Shah *et al.* (2017).

The data in Table 2 reveal that the mean neutrophils count was significantly increased (76.50 ± 2.66 vs. 64.25 ± 1.90 %), whereas the values of lymphocytes, monocytes and eosinophils were decreased in bitches with pyometra as compared to healthy ones. The values of neutrophil count in bitches with pyometra decreased gradually and significantly on day 8th and 15th post-hysterectomy, whereas in normal healthy bitches the values were statistically similar. The mean lymphocytes count in bitches with pyometra was significantly lower than in normal healthy bitches on day 0 (20.70 ± 2.49 vs. 29.63 ± 2.38 %), and it increased gradually and significantly by 8th and 15th day post-hysterectomy, whereas no such change was noted in normal healthy bitches after spaying. These observations on DLC compared well with the reports of Dabhi *et al*. (2009), Gupta and Dhami (2013), Patil *et al*. (2013), Mahesh *et al*. (2014) and Shah *et al*. (2017) in which they noted increased level of PMN cells mostly neutrophils due to toxemic condition.

Biochemical Alterations

It is evident from the Table 3 that there was significant increase in values of plasma urea nitrogen (46.13 ± 6.79 vs. 24.32 ± 5.16 mg/dl), plasma total protein (7.30 ± 0.26 vs. 5.56 ± 0.29 g/dl) and globulin (4.87 ± 0.27 vs. 2.85 ± 0.19 g/dl) with lower A:G ratio (0.51 ± 0.03 vs. 0.99 ± 0.10) in bitches with pyometra as compared to healthy ones particularly before operation. There was decline in the values of plasma urea nitrogen and creatinine during post-operative period, yet the variations in the values of all these constituents were non-significant between periods in pyometra affected and healthy bitches. The plasma levels of aspartate amino transferase, alanine aminotransferase and total cholesterol were apparently much higher in bitches with pyometra than the healthy ones, but did not differ statistically except AST, and the values at 8th and 15th day post-operative, decreased significantly in both healthy and affected bitches (Table 3). Statistically non-significant differences found in levels of these constituents could be attributed to huge variation in the values among bitches affected with open and closed pyometra. These findings of biochemical analysis to some extent corroborated with the earlier reports of Dabhi *et al.* (2007), Murthy *et al.* (2013), Dabrowski and Wawron (2014), and Prasad *et al*. (2017). These authors stated that the levels of plasma urea nitrogen, plasma creatinine, ALT, AST were higher in bitches affected with pyometra. Wheaton *et al.* (1989) opined increased levels of BUN and creatinine in pyometra due to decreased blood flow to kidneys.

Conclusion

The study revealed that haematological values such as Hb, PCV, TEC, TPC and TLC could be used as good indicator for the diagnosis of pyometra. These values were significantly higher and were found to improve after ovario-hysterectomy. A marked leucocytosis (37,000/ cmm) and neutrophilia (79.16 %) was observed particularly in closed pyometra cases which were declined post-operatively. The significantly increased plasma urea nitrogen, creatinine, total protein, globulin, total cholesterol and enzymes AST-ALT profile reflected vital organ damage from pyometra/toxaemia, which could be reverted back to normal following ovario-hystertectomy.

Acknowledgement

We thank the Dean, College of Veterinary Science and Animal Husbandry, Anand, and professor & Head, Veterinary Clinical Complex and Veterinary Surgery for providing funds and necessary facilities to undertake this study.

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