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# Pyometra in bitches: A haematological, biochemical and ultrasonographical study

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#### **Abstract**

Ten no. of non-pregnant bitches in diestrous phase (control group /Group I) and 10 nos of bitches showing symptoms of pyometra (Group II) were selected for study of haematological, biochemical and haemodynamics parameters. A highly significant difference (P< 0.05) was observed in the PCV and TLC values between normal dioestrus and pyometric bitches abut there is no significant difference in the haemoglobin level and TEC values. A significant (P< 0.05) increase in the mean of BUN, Creatinine and ALP level was observed in pyometric bitches as compared to control group. Control group bitches showed higher hemodynamic indices (PI & RI) and lower blood flow velocities (PSV and EDV) in comparison to pyometra group. As the blood flow in the uterine artery increases significantly during pyometra, these parameters can be used as a key parameter for diagnosis as well as therapeutic evaluation in canine pyometra.

Keywords: Haematology, biochemical, ultrasound, pyometra

#### Introduction

The cystic endometrial hyperplasia-pyometra complex (CEH-P) is one of the most prevalent uterine diseases in the female dog. Middle aged to old, diestrous bitches are frequently affected (Jena *et al.*, 2013) [12]. Cystic endometrial hyperplasia is a subclinical disease characterized by the proliferation of endometrial glands resulting in the formation of fluid-filled cysts and luminal uterine contents which may be present (Batista *et al.*, 2013) [1]. Ultrasonographic characteristics of pyometra vary depending upon the extent of involvement and nature of content. Extensive involvement depicts round hypoechoic to anechoic areas placed side by side covering the complete abdomen while moderate involvement exhibits hypoechoic, roughly round structure ventral to ventro-lateral to the anechoic urinary bladder in transverse section Mild involvement is more readily visualised on longitudinal section as mixed anechoic to hypoechoic tubular structure (Singh *et al.*, 2010) [21].

### **Materials and Methods**

The Research work was carried out at TVCC in collaboration with the Department of Animal Reproduction, Gynaecology and Obstetrics, College of Veterinary Science & A.H., OUAT, Bhubaneswar. Ten no. of non-pregnant bitches in diestrous phase (control group /Group I) were selected along with 10 nos of bitches showing symptoms of pyometra (Group II). Relevant haematological test like i.e. Total Leukocyte Count (TLC), Total Erythrocyte Count (TEC), Haemoglobin (Hb) and Packed Cell Volume (PCV) and Biochemical test like i.e. (BUN, Creatinine and ALP) were performed in pyometric cases and compared with control group. Two dimensional ultrasonographic evaluations of the uterus was carried out by using (3 -11 MHz) convex probe of a real time doppler ultrasonography machine (ALOKA PROSOUND ALPHA 6). The widest cross sectional area of the uterus was taken using Bmode ultrasonography. Then colour doppler was conducted to localize uterine arteries at both sides of the uterine body and pulse wave Doppler was performed to obtain the waveform. The angle of insonation was adjusted manually and measurements with an angle less than 30° were only considered for analysis. Three consecutive waveforms with maximum Doppler shift included in the study. Peak systolic Velocity (PSV; cm/s), End diastolic Velocity (EDV; cm/s), Resistive Index (RI) and Pulsatility Index (PI) were measured automatically.

#### **Results & Discussion**

The mean values of haemoglobin (g/dl), Packed cell Volume (PCV %), Total erythrocyte count (TEC x 10<sup>6</sup>/mm<sup>3</sup>) and Total leukocyte Count (TLCx 10<sup>3</sup>/UL) has been recorded as 12.33

 $\pm$  0.33, 40.87  $\pm$  0.92, 6.68  $\pm$  0.17 and 14.31  $\pm$  2.15 respectively in Gr. I bitches (Table 1). The corresponding mean values were  $11.03 \pm 0.27$ ,  $34.29 \pm 0.86$ ,  $5.53 \pm 0.22$  and  $35.59 \pm 8.32$  in Gr. II. Statistical analysis indicated that there was highly significant difference (P< 0.05) in the PCV and TLC values between normal dioestrus and pyometric bitches abut there is no significant difference in the haemoglobin level and TEC values. In the present study in normal dioestrus bitches haemoglobin level ranges between 12-13gm/dl. However the mean haemoglobin level was decreased in bitches affected with pyometra indicating anemia which is in agreement with the findings of Hagman et al. (2009) [9], Nath et al (2009a) [14] Jena et al. (2013) [12] and samantha et al (2018) [17]. This might be due to loss of red blood cells by diapedesis into uterine lumen apart from depressed feed intake and impaired erythropoiesis under toxemic condition in severely affected cases reported by Dabhi et al. (2009) [4] and Jena et al. (2013) [12]. However, in the present study most of the pyometra affected bitches showed haemoglobin level within normal range which is similar to the finding of Schepper et al. (1987) [18] which might be due to concurrent dehydration as reported by Huszenickza et al. (1985) [11]. In contrary Bigliardi et al. (2004) [3] reported that Hb level was within the normal range in 100 per cent of the pyometra affected bitches. In the present study the mean PCV level was decreased in bitches affected with pyometra as compared to normal dioestrus bitches indicating a indicating a mild normocytic, normochromic and regenerative type of anaemia which is in agreement with the findings of Dabhi et al. (2009) [4] and Jena et al. (2013) [12] which might be due to concomitant dehydration as reported by Verstegen et al. (2008) [23]. In contrary to the above reports, Bigliardi et al. (2004) [3] reported no change in the PCV level in pyometra. In the present study the mean total erythrocytic count in bitches affected with pyometra is less as compared to normal dioestrus bitches, which was in agreement with the findings of Dabhi et al. (2009) [4] and Nath et al. (2009a) [14]. According to the reports of Schepper et al. (1987) [18] and Wheaton et al. (1989) [24], normocytic, normochromic anaemia might be associated with the toxic depression of the bone marrow whereas severe non-regenerative, microcytic, hypochromic anaemia accompanied by extremely high white blood cell levels might be indicative of a concurrent blood loss possibly by diapedesis into the luminal pus and due to shortened life span of circulating erythrocytes associated with iron deficiency. In contrary to the above reports, Bigliardi et al. (2004) [3] reported no change in TEC level in pyometra. In the present study, leucocytosis were the most consistent finding among the bitches affected with pyometra which was in agreement with the reports of Dabhi et al. (2009) [4] and Nath et al. (2009a) [14] and Jena et al. (2013) [12]. This might be due to increased stress on the body defense mechanism which in turn produced increased leucocytes to combat the infection as reported by Nath et al. (2009a) [14]. Different degree of leucocytosis was observed in bitches affected with pyometra might be due to severity of the inflammation varying between animals as reported by Dabhi et al. (2009) [4]. In contrary to the above reports, Nelson and Feldman (1986) [16] reported that a normal leucogram occurred in pyometra.

The mean values of serum BUN (mg/dl), Creatinine (mg/dl) and ALP (mg/dl) have been recorded as  $26.27\pm1.47$ ,  $1.30\pm0.09$  and  $121.28\pm4.77$  respectively in normal dioestrus bitches. The corresponding mean values are  $32.89\pm1.18$ ,  $2.21\pm0.07$  and  $158\pm7.63$  respectively in pyometric bitches.

Statistical analysis revealed that there is a significant (P< 0.05) increase in the mean of BUN, Creatinine and ALP level in pyometric bitches as compared to control group. In the present study, the mean BUN and creatinine level in serum were found to be elevated in bitches affected with pyometra as compared to normal dioestrus bitches which were in agreement with the findings of Bigliardi et al. (2004) [3]. Elevated BUN and creatinine level might be due to toxaemia, in turn leading to decomposition of body protein as a result of suppurative process and reduced renal perfusion and dehydration leading to immune complex deposition in the glomeruli causing glomerulonephritis and proximal tubular damage resulting in renal failure depending upon the level of toxaemia and dehydration caused by pyometra as reported by Gayakawad et al. (1999) [7] and Nath et al. (2009b) [15]. However, Nak et al. (2004) [13] reported that pyometra might cause renal failure due to the effect of bacterial toxins especially, Escherichia coli on renal tubules. Singh et al. (2006) [20] reported that an increased level of BUN in pyometric bitches might be associated with an increased production or a faulty elimination of urea. Faulty elimination of urea in urine was associated with either pre renal or renal factors. The pre renal factors like haemoconcentration or low blood pressure might lead to a lowering of the glomerular filtration rate and impair the efficiency of the kidney. In contrary, Hagman et al. (2006) [8] and Hagman et al. (2009) [9] reported that BUN and creatinine values of the affected bitches did not differ significantly from normal values demonstrating a normal kidney and liver function and lack of hepatocellular damage in the most dogs. Verstegen et al. (2008) [23] reported that serum blood urea nitrogen and creatinine concentrations were not usually elevated, unless pre-renal azotemia developed as a consequence of dehydration. Hagman et al. (2009) [9] and Singh et al. (2006) [20] reported that the levels of creatinine did not differ significantly in comparison with healthy dogs. The mean alkaline phosphatase level in serum were found to be elevated in all bitches affected with pyometra as compared to normal dioestrus bitches which were in agreement with the findings of England et al. (2007) [5] which might be due to intrahepatic cholestasis as reported by Hagman et al. (2009) [9]. However, Verstegen et al. (2008) [23] reported that an mild to moderate elevation in ALP in bitches affected with pyometra was observed that might be due to hepatocellular damage caused by septicaemia and endotoxaemia, diminished hepatic circulation and cellular hypoxia due to dehydration in the bitches. Sevelius et al. (1990) [19] reported elevated SAP levels above the normal range in more than 50 per cent of bitches affected with pyometra and also stated that the elevated SAP activity originating from liver tissue was a common finding in inflammatory and infectious conditions.

The mean value of PI in control group and pyometric bitches has been recorded as  $1.296\pm0.017$ , and  $1.15\pm0.013$  respectively. PI values of control group bitches ranges between 1.21-1.37 and in case of pyometra between 1.11-1.24. PI values varied significantly (P<0.01) between control group and pyometric bitches. The mean value of RI in control group and pyometric bitches has been recorded as  $0.634\pm0.027$  and  $0.246\pm0.016$ . RI values of control group bitches ranged from 0.42-0.72 and in pyometric bitches between 0.20-0.33. There is significant difference in the RI values between these two groups (P<0.01). The mean values of PSV in control group and pyometric bitches has been recorded as  $0.140\pm0.161$ , 0.161

values were  $1.819 \pm 0.262$  and  $27.570 \pm 0.452$ . PSV values of control group bitches ranges between 2.70 - 11.99 cm/s and in pyometric bitches 67.30 - 72.12 cm/s. Similarly EDV values of control group bitches ranges between 0.87 - 3.28cm/s and in pyometric bitches 25.43 – 29.56 cm/s. Statistical analysis revealed that there is high significant difference (P< 0.01) level in the PSV and EDV values of control group and pyometric bitches. In the present study pyometra affected bitches showed significantly higher resistance Doppler wave form of uterine arteries as compared to control group. RI values found in case of control group bitches is higher than that of pyometric group bitches which is similar to as reported by Batista et al. (2016) [2]. Control group bitches (PSV:  $6.140\pm1.161$ ; EDV: 1,819  $\pm0.262$ ) showed lower blood flow velocities as compared to pyometra groups (PSV: 69.006 ± 0.473; EDV:  $27.570 \pm 0.452$ ). Conversly, Control group bitches (RI: 0.634±0.027; PI: 1.296±0.017) showed higher hemodynamic indices in comparison to pyometra groups (RI:  $0.246 \pm 0.016$ ; PI:  $1.15 \pm 0.013$ ) which is similar to the findings of (Gal et al., 2012) [6]. Increased blood flow, vasodialtion and angiogenesis are the common findings during any inflammatory process. Heap et al. (1975) [10] and Still et al. (1978) [22] reported that higher intrauterine prostaglandin E concentration may lead to an increase in the uterine perfusion. During endometrial inflammation, several mammalian species locally release nitric oxide as potent vasodilator in addition to prostaglandin.

#### Conclusion

The PI, RI, PSV and EDV values varied significantly between pyometra and control group bitches. There was significant alteration in haematological and biochemical parameters except haemoglobin and total erythrocytic count in pyometra affected bitches compared to healthy bitches. The blood flow in the uterine artery increases significantly during pyometra which can be used as a key parameter for diagnosis as well as therapeutic evaluation in canine pyometra.

**Table 1:** Haematological, Biochemical and haemodynamics changes during pyometra in bitches

| Parameters        | GR-I                 | GR-II                      |
|-------------------|----------------------|----------------------------|
| Hb(gm/dl)         | 12.33°± 0.33         | $11.03^a \pm 0.27$         |
| PCV (%)           | $40.87^{a}\pm0.92$   | $34.29^{b} \pm 0.86$       |
| TECX106/UL        | 6.68 a± 0.17         | $5.53^{a} \pm 0.22$        |
| TLC(X103/UL)      | $14.31^a \pm 2.15$   | $35.59^{b} \pm 8.32$       |
| BUN(mg/dl)        | $26.27^{a} \pm 1.47$ | 32.89 b±1.18               |
| Creatinine(mg/dl) | $1.30^{a}\pm0.09$    | $2.21^{b} \pm 0.07$        |
| ALP (mg/dl)       | $121.28^a \pm 4.77$  | $158.59^{b} \pm 7.63$      |
| PI                | 1.296a±0.017         | 1.15 <sup>b</sup> ±0.013   |
| RI                | 0.634a±0.027         | 0.246 <sup>b</sup> ±0.016  |
| PSV               | 6.140a±1.161         | 69.006 <sup>b</sup> ±0.473 |
| EDV               | 1.819a±0.262         | 27.570 <sup>b</sup> ±0.452 |

Means bearing different superscripts between columns differ significantly (p<0.05)

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