Comparative studies on changes in C-reactive protein, serum cortisol, blood glucose and aspartate amino transferase level following left flank method and laparoscopic method of ovariohysterectomy in bitches

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ABSTRACT

A study was carried in 12 bitches presented to Veterinary College Hospital, Bangalore, for elective ovariohysterectomy. They were divided into group A and group B, with six bitches each. Bitches of Group A were sterilized by conventional method of ovariohysterectomy by left flank approach, and Group B bitches were sterilized by laparoscopic method. Blood samples were collected at regular intervals to estimate C-reactive protein (CRP), serum cortisol, blood glucose and aspartate amino transferase (AST) level. Significant elevation of CRP, blood glucose and AST levels were noticed in bitches of Group A compared to laparoscopic Group B. Serum cortisol level was elevated for longer period of time in Group A bitches compared to Group B. This study suggests that laparoscopic method of ovariohysterectomy causes less tissue damage, trauma and post-operative pain than conventional method of ovariohysterectomy in bitches.

Key words: laparoscopic ovariohysterectomy, C-reactive protein, serum cortisol, blood glucose, aspartate amino transferase

Introduction

Ovariohysterectomy is the most common surgical procedure performed in Veterinary practice for sterilization in dogs (BLOOMBERG, 1996). Ovariohysterectomy prevents or reduces the risk of development of mammary tumour, pyometra, the inconvenience of vaginal discharge and male attraction during estrus (DAVIDSON et al., 2004). In
traditional ovariohysterectomy the uterus and ovaries are double ligated, transected and abdomen is typically closed in three layers (FINGLAND, 1998). Laparoscopic method of ovariohysterectomy is an alternative to traditional abdominal ovariohysterectomy in bitches, having advantages like lower risk of dehiscence, early wound healing and shortened hospitalization (DAVIDSON et al., 2004). Laparoscopic method of ovariohysterectomy causes less post-operative pain compared to conventional method of ovariohysterectomy in bitches (DEVITT et al., 2005). The present study was carried out to compare tissue damage, post-operative stress, pain and wound healing following conventional and laparoscopic method of ovariohysterectomy in bitches.

Materials and methods

Twelve healthy bitches presented to Department of Surgery and Radiology, Veterinary College Hospital, Hebbal, Bangalore, for elective ovariohysterectomy ranging in body mass from 12 to 18 kg and 1 to 2 years of age were included for this study. All the bitches were subjected to clinical examination to rule out pregnancy and hematobiochemical tests to assess their fitness for the surgery. Selected bitches were randomly divided into two groups Viz., Group A and B of six animals each. All the bitches were premedicated with diazepam* 0.5 mg/kg body mass intravenously. After 15 minutes, general anesthesia was induced and maintained with thiopentone sodium** (2.5%) given to the effect at the dose rate of 25 mg/kg body mass intravenously. (*Calmpose inj., 2 mL amp, Ranbaxy Laboratories Ltd, Mumbai, India; **Thiosol inj., 500 mg vial, Neon Laboratories Ltd, Mumbai, India.)

Group A bitches were sterilized by left flank method of ovariohysterectomy. Group B bitches were sterilized by laparoscopic method of ovariohysterectomy. All the Group B bitches were positioned in dorsal recumbency with elevated pelvis (30˚), all the limbs were secured separately and ribcage was supported by sand bags. Carbon dioxide gas was insufflated into the abdomen to an intra-abdominal pressure of 12-14 mm Hg. Median port was made by introducing 10 mm trocar at the umbilicus into the abdomen with the tip of trocar directing dorsally to avoid injury to visceral organs. Two more paramedian ports were made approximately five cm caudolateral to median port on either side by inserting five mm trocars. Laparoscope was inserted into median port and paramedian ports were used as the instrument port. Hemostasis was achieved by bipolar electrocautery. After complete separation, the genital tract was removed via median port.

Blood samples were collected in Ethylene Diamine Tetra-acetic Acid (EDTA) vials and serum vials pre-operatively, immediately after operation, and 4, 8, 24, 48 and 72 hours post-operatively for estimation of CRP, serum cortisol and blood glucose. CRP (mg/L) was estimated by turbidometric assay using blood plasma (KJELGAARD et al., 2004) using M/s Agappe diagnostics, Kerala, India. Serum cortisol (µg/dL) was measured by two-
side sandwich chemiluminescent immuno assay at Thyrocare technologies Ltd., Thane, Mumbai, India. Serum samples were stored and transported at -20 °C till estimation. Blood glucose was estimated by standard method using blood plasma (HENRY, 1979) by way of the M/s Swemed diagnostic kit, Bangalore, India. Blood samples were collected in EDTA vials pre-operatively, and 6, 12, 24, 48, 72 hours and 7 days post-operatively for estimation of AST. Aspartate amino transferase was estimated by standard method using blood plasma (HENRY, 1979), by way of the M/s Swemed diagnostic kit, Bangalore, India.

Results

The mean CRP level ranged from 3.00 ± 0.77 to 8.83 ± 1.17 in Group A bitches, and 2.67 ± 0.92 to 5.67 ± 1.48 in Group B bitches respectively (Table 1). Statistically there was significant elevation (P≤0.05) in mean CRP level in Group A compared to Group B between 24 hours to 48 hours post-operatively. The mean serum cortisol level ranged from 1.18 ± 0.25 to 5.33 ± 0.52 in Group A bitches, and from 0.70 ± 0.27 to 5.27 ± 0.73 in Group B bitches (Table 1). Serum cortisol levels were significantly elevated (P≤0.05) in Group A bitches up to 4 hours after operation compared to Group B bitches.

Table 1. Mean ± SE values of serum cortisol, blood glucose and C - reactive protein in group A and B bitches

<table>
<thead>
<tr>
<th>Time interval</th>
<th>Serum cortisol (μg/dL)</th>
<th>Blood glucose (mg/dL)</th>
<th>C-reactive protein (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A</td>
<td>Group B</td>
<td>Group A</td>
</tr>
<tr>
<td>Before surgery</td>
<td>2.15 ± 0.56</td>
<td>2.45 ± 0.58</td>
<td>32.83 ± 8.12</td>
</tr>
<tr>
<td>After surgery</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immediate</td>
<td>5.33 ± 0.52&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.27 ± 0.73&lt;sup&gt;b&lt;/sup&gt;</td>
<td>95.50 ± 7.54&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>4 H</td>
<td>5.20 ± 0.94&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.02 ± 1.07</td>
<td>33.00 ± 6.14</td>
</tr>
<tr>
<td>8 H</td>
<td>3.17 ± 0.48</td>
<td>3.42 ± 0.90</td>
<td>45.00 ± 10.19</td>
</tr>
<tr>
<td>24 H</td>
<td>1.48 ± 0.53</td>
<td>1.28 ± 0.23</td>
<td>65.67 ± 17.75</td>
</tr>
<tr>
<td>48 H</td>
<td>1.18 ± 0.25</td>
<td>0.70 ± 0.27</td>
<td>54.00 ± 16.17</td>
</tr>
<tr>
<td>72 H</td>
<td>1.33 ± 0.26</td>
<td>1.67 ± 0.45</td>
<td>55.00 ± 10.53</td>
</tr>
</tbody>
</table>

Group A - surgical method of ovariohysterectomy. Group B - laparoscopic method of ovariohysterectomy. Mean ± SE values with superscripts (a, b) vary significantly (P≤0.05) between Group A and B. Mean ± SE values with superscript (x) vary significantly (P≤0.05) within the group.
Table 2. Mean ± SE values of aspartate amino transferase levels in group A and B bitches

<table>
<thead>
<tr>
<th>Aspartate amino transferase (IU/L)</th>
<th>Group A</th>
<th>Group B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before surgery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>35.67 ± 1.82</td>
<td>35.50 ± 2.26</td>
<td></td>
</tr>
<tr>
<td>After surgery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>36.67 ± 0.99</td>
<td>33.83 ± 3.84</td>
<td></td>
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<tr>
<td>43.50 ± 1.65*</td>
<td>40.67 ± 2.40</td>
<td></td>
</tr>
<tr>
<td>52.50 ± 4.54*</td>
<td>43.33 ± 2.38</td>
<td></td>
</tr>
<tr>
<td>55.67 ± 1.43**</td>
<td>41.83 ± 1.21</td>
<td></td>
</tr>
<tr>
<td>53.33 ± 4.09**</td>
<td>41.00 ± 1.92</td>
<td></td>
</tr>
<tr>
<td>40.83 ± 3.35</td>
<td>37.33 ± 2.38</td>
<td></td>
</tr>
</tbody>
</table>

Group A - surgical method of ovariohysterectomy. Group B - laparoscopic method of ovariohysterectomy. Mean ± SE values with superscripts (a, b) vary significantly (P≤0.05) between Group A and B. Mean ± SE values with superscript (x) vary significantly (P≤0.05) within the group.

The mean blood glucose level ranged from 32.83 ± 8.12 to 95.50 ± 7.54 in Group A bitches and 28.00 ± 7.91 to 75.17 ± 8.16 in Group B bitches. (Table 1). Statistically there was significant elevation (P≤0.05) in mean blood glucose level in the Group A compared to Group B during immediate postoperative period. The mean AST level ranged from 35.67 ± 1.82 to 55.67 ± 1.43 in Group A bitches, and from 33.83 ± 3.84 to 43.33 ± 2.38 in Group B bitches (Table 2). There was significant elevation (P≤0.05) in the mean AST level in Group A compared to Group B bitches between 48 to 72 hours post-operatively.

**Discussion**

C-reactive protein is the fastest reacting canine acute-phase protein, which increases in response to infection and tissue injury (CONNER et al., 1998). There was a significant elevation in C-reactive protein level at 24 hours to 48 hours after operation in Group A compared to Group B bitches. This could be due to a greater tissue damage and inflammatory phenomenon in Group A bitches than in Group B bitches (CONNER et al., 1998). RANGANATH and SENTHIL (2006) reported that C-reactive protein level peaked at 24 hours following surgical method of ovariohysterectomy in mongrel bitches. Assay of cortisol concentration has been used as an indicator of stress and pain in dogs (HANSEN et al., 1997; DEVITT et al., 2005; HANCOCK et al., 2005; MALM et al., 2005), in cats (SMITH et al., 1996), in lambs (SHUTT et al., 1987), in pigs (PARROTT et al., 1989) and in human
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beings (BASSETT et al., 1987). Significant rise in the serum cortisol level for longer period in Group A bitches compared to Group B bitches suggests that surgical method of ovariohysterectomy is more stressful and painful in bitches than laparoscopic method (DEVITT et al., 2005).

Significantly higher blood glucose level was noticed in Group A compared to B Group bitches during immediate post-operative period. DEVITT et al. (2005) also observed significant increase in blood glucose concentration at 1, 2, 4 and 6 hours following an ovariohysterectomy, and at one hour following laparoscopic ovariohysterectomy in bitches.

Elevated blood glucose level in Group A bitches could be due to stress, pain and increased cortisol level (BENJAMIN, 2001). Significant elevation of AST levels were noticed in Group A bitches compared to Group B bitches during 48 to 72 hours post-operatively. This could be attributed to excess muscle trauma (KANEKO, 1980). SCHMIDT and BOOKER (1982) reported significant increase in AST level up to 72 hours after operation. This could be due to excess muscle damage in Group A bitches compared to Group B bitches. In conclusion, the laparoscopic method of ovariohysterectomy proved to be a better technique compared to the conventional method of ovariohysterectomy in bitches, as the procedure caused less tissue damage, inflammation, post-operative stress and pain.

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SAŽETAK
Istraživanje je provedeno na 12 kuja podijeljenih u dvije skupine po 6 kuja. Prva skupina bila je podvrgnuta laparotomiji u lijevim slabinama i klasičnoj ovariohisterektomiji dok je druga skupina operirana laparoskopski. Poslije operacije redovito je uzimana krv radi praćenja razine C-reaktivnog proteina, serumskog kortizola, glukoze u krvi i aspartat aminotransferaze (AST). U skupini podvrgnutoj klasičnoj ovariohisterektomiji uočeno je signifikantno povećanje razine C-reaktivnog proteina, serumskog kortizola, glukoze u krvi i AST. Razina serumskog kortizola bila je povećana duže razdoblje u skupini operiranoj klasičnim postupkom ovariohisterektomije. Rezultati studije upućuju na zaključak da laparoskopska metoda uzrokuje slabije oštećenje tkiva i manju postoperativnu bol u usporedbi s klasičnom ovariohisterektomijom.

Ključne riječi: laparoskopska ovariohisterektomija, C-reaktivni protein, serumski kortizol, glukoza u krvi, aspartat aminotransferaza