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Research Article

COMPARATIVE STUDIES ON STRESS RESPONSE TO LAPROSCOPIC OVARIECTOMY AND OPEN OVARIECTOMY IN DOGS

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ABSTRACT

Twelve clinically healthy animals (aged between one to three years and weighing between 12-24 kilograms body weight) brought for sterilization in Veterinary clinical complex, LUVAS, Hisar were included in this study. All the dogs were randomly divided into two groups; dogs in group I, were sterilized by open ovariectomy whereas dogs of group II, were subjected to laparoscopic ovariectomy. Surgical anesthesia was achieved by using a combination of xylazine-ketamine @ 1 mg/kg body weight and 5mg/kg body weight, respectively by intramuscular route in dogs premedicated with atropine sulphate @ 0.02 mg/kg b.wt by intramuscular route. Maintenance of anesthesia, was done by xylazine-ketamine combination @ 0.5 mg/kg b.wt and 2.5 mg/kg b.wt, respectively by IV. Blood samples were collected at regular intervals to estimate aspartate amino transferase, creatine kinase, C-reactive protein, cortisol and glucose. Recording of pain score was done using numerical rating scale (NRS). Significant changes between the groups were also seen in pain score, aspartate amino transferase, creatine kinase, C-reactive protein, serum cortisol and glucose. The result of this study indicated that laparoscopic ovariectomy is less painful, less stressful and less traumatic in comparison to open ovariectomy.

Keywords: Dogs, Laparoscopic, Ovariectomy, Stress

Pet population control continues to be the cause of social and economic importance. Although pharmacologic approaches to the fertility control continue to be evaluated clinically for the use on an individual basis, the need remains for an effective, rapid, safe and economic means of sterilizing pets adopted by human societies or pet shelter programmes (Wildt and Lawler, 1985). In recent years, there has been much interest in minimally invasive methods (Mayhew, 2011), including laparoscopic techniques e.g. laparoscopic ovariectomy (LapOVE). Since these are thought to be more appropriate for 'out-patient settings' in small animal practice because the smaller incisions used are less painful than those used in traditional approaches and result in less surgical trauma, faster recovery and less patient immobilisation (Davidson *et al.*, 2004; Devitt *et al.*, 2005; Hancock *et al.*, 2005; Culp *et al.*, 2009). Laparoscopic surgery has been one of the fastest growing areas in modern surgery. However, in veterinary medicine, the field of MIS (Minimum invasive surgery) is still very much in its infancy and further evidence-based randomised studies are required (Mayhew, 2011).

In the view of above, the study was conducted to compare tissue damage, post-operative stress, pain and wound healing following open ovariectomy and laparoscopic ovariectomy in dogs.

MATERIALS AND METHODS

Twelve clinically healthy animals (aged between one to three years and weighing between 12-24 kilograms body weight) brought for sterilization in Veterinary Clinical Complex, LUVAS, Hisar were encompassed in this study. All the dogs were randomly divided into two group viz., group I and group II. Surgical anesthesia was achieved by using a combination of xylazine-ketamine @ 1 mg/kg b.wt. and 5 mg/kg b.wt., respectively intramuscularly in dogs premedicated with atropine sulphate @ 0.02 mg/kg b.wt. intramuscularly. Maintenance of anesthesia was done by xylazine-ketamine combination @ 0.5 mg/kg b.wt. and 2.5 mg/kg b.wt., respectively by i/v route.

Group I dogs were sterilized by traditional open ovariectomy, whereas group II dogs were sterilized by laparoscopic ovariectomy. In group I, a midline skin incision was given from the umbilicus and extended 3-4 cm caudally. The ovaries were identified following either the left or right uterine horn proximally and two simple ligatures were placed around the ovarian artery and vein. The mesovarium and proper ligament were transected and the ovary was removed. The ovarian artery and vein were then ligated and severed at the proper ligament. In this way, cranial tip of the uterine horn was ligated and the ovary was removed. The stump was checked for hemorrhage and released in the abdomen. Finally the incision was closed in a routine three- layer manner. For laparoscopic ovariectomy, all the animals were positioned in dorsal recumbency and two-port LapOVE technique was used. Using the left hand, the abdominal wall on the umbilical area was grasped and elevated and veress needle was introduced in the abdomen. Then pneumoperitoneum was established by connecting the needle to the high flow insufflators using carbon dioxide at a pressure of 12 mm Hg. Using blade no. 11, a 5

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mm stab incision was made 2 cm caudal to umbilicus through the skin and subcutaneous tissue down to the linea alba and non-threaded cannula-trocar assembly was inserted into the abdomen through the stab incision. The trocar was then removed and the 30º camera (Karl Storz, Tuttlingen, Germany) connected to a light source was inserted into the abdomen and a 360º scan was performed to check for any existing abnormalities. A 5 mm skin incision was made cranial to the umbilicus at a point midway between xiphoid process and the umbilicus and the second 5 mm threaded cannula was inserted under direct visualization to prevent injury to abdominal organs. The dogs were tilted 30º either to the right or left lateral recumbency to perform left or right OVE, respectively. For OVE, the left or right kidney was identified as a land mark. The right ovary was found lying on the caudal pole of the right kidney and 5 mm grasping forceps was passed through the cranial port and was directed towards the caudal pole of the right kidney. The ovariouterine junction was grasped and elevated by grasping forceps and was tacked to theventro-lateral abdominal body wall by passing a 5 cm, 3/8 circle curved cutting swaged needle and was sutured percutaneously through the body wall. Under the observation of laproscope, the needle was passed directly into the abdomen from lateral to medial direction through the mesovarium. Then, the 5 mm bipolar electrocautery forceps was introduced via cranial port and the ovarian pedicle, proper ligament and mesovarium were cauterized. Following haemostasis of the ovarian pedicle, the 5 mm laparoscopic scissors was inserted into abdomen from the cranial portal to resect the ovary.

At the end of the procedure, the grasping forceps was introduced for holding the resected ovary. Once the ovary was grasped, the needle holding the tacking suture was released and the forceps holding the ovary was pulled backwards until the forceps entered inside the cannula under the direct visualization of the laparoscope. It may be necessary to slightly enlarge the portal openings to allow the ovary to be removed. To avoid losing the ovary, an artery forceps was used to grasp the ovary as it exits the abdomen. The patient was tilted 30º to the other side to perform OVE on the contralateral side. The same procedure was repeated to remove the left ovary. After removing the ovaries, the abdomen was scanned for ensuring hemostasis or any other complications. The portal sites were sutured using monofilament synthetic suture material USP-1 for muscles and for skin, non absorbable multifilament USP-1 suture material was used in interrupted horizontal mattress pattern.

Recording of pain score was done preoperatively (T1), after 15 minutes from anaesthesia recovery (T2), 4h (T3), 8h (T4), 24h (T5) and post operatively after 48h (T6) using numerical rating scale. Blood samples were collected in serum vials pre-operatively (T1), after 15 minutes recovery from anaesthesia (T2), and 4h (T3), 8h (T4), 24h (T5), 48h (T6) and 7th day post-surgery (T7) post-operatively for estimation of aspartate amino transferase, creatinekinase, C-reactive protein, serum cortisol and blood glucose. Biochemical examinations were performed using commercially available kits (Transasia Bio Medical Ltd, India). Glucose was estimated from plasma collected in 3.8% sodium fluoride.

RESULTS AND DISCUSSION

A significantly lower ($p< 0.05$) pain score values were recorded at 8h, 24h and 48h post operatively in laparoscopic ovariectomy group in comparison to the values recorded in open ovariectomy group. Significantly higher pain score values in group I may be due to more tissue trauma encountered during open method in comparison to laparoscopic method.

Table 1

Comparative NRS, AST and CK values at different time intervals in ovariectomy in dogs (Mean±S.E.)

Time Interval	Pain Score (n=20)		Aspartate Amino Transferase (IU/L)		Creatine Kinase (U/L)	
	Group I	Group II	Group I	Group II	Group I	Group II
TI	0	0	35.66 ± 0.34^{a}	35.33 ± 0.16^{a}	208.46 ± 0.81^{a}	202.45 ± 1.21^{b}
T2	8.50 ±0.56^{a}	7.50 ± 0.42^{a}	36.66 ± 0.34^{a}	33.65 ± 0.16^{b}	259.88± 1.15^{a}	252.53 ± 1.23^{b}
T3	9.56 ±0.71^{a}	9.33 ± 0.33^{a}	43.51 ± 0.34^{a}	40.95 ± 0.16^{b}	440.34 ± 0.80^{a}	441.31 ± 1.19^{b}
T4	9.00 ± 0.85^{a}	6.83 ± 0.30^{b}	52.50 ±0.34^{a}	43.15 ± 0.16^{b}	675.17 ± 0.82^{a}	651.47 ± 1.30^{b}
T5	9.50 ± 0.84^{a}	5.83 ± 0.16^{b}	55.31 ± 0.20^{a}	41.67 ± 0.17^{b}	593.04 ± 0.67^{a}	581.95 ± 1.32^{b}
T6	8.33 ± 0.49^{a}	3.00 ± 0.44^{b}	53.33 ± 0.34^{a}	41.01 ± 0.17^{b}	544.53 ± 2.94^{a}	520.74 ± 0.67^{b}
T7	—	—	40.82 ± 0.34^{a}	37.15 ± 0.16^{b}	436.59 ± 1.04^{a}	410.15 ± 1.08^{b}

Values within the group with different superscripts differ significantly ($p< 0.05$)

T1: 1h before surgery, T2: 15 minutes after recovery from anaesthesia, T3: 4h after surgery, T4: 8h after surgery, T5: 24h after surgery, T6: 48h after surgery, T7: 7th day of surgery.

Table 2

Comparative CRM, cortisol and glucose values in ovariectomy in dogs (Mean±S.E.)

Time Interval	C-Reactive Protein		Serum Cortisol (µg/ml)		Plasma Glucose (mg/ml)	
	Group I	Group II	Group I	Group II	Group I	Group II
TI	3.32 ± 0.02^{a}	2.61 ± 0.04^{b}	2.16 ± 0.03^{a}	2.40 ± 0.02^{b}	32.82 ± 0.02^{a}	38.25 ± 0.23^{a}
T2	3.75 ± 0.03^{a}	3.01 ± 0.02^{b}	5.32 ± 0.02^{a}	5.21 ± 0.02^{b}	95.58 ± 0.02^{a}	74.99 ± 0.16^{b}
T3	4.50 ± 0.02^{a}	3.93 ± 0.02^{b}	5.20 ± 0.02^{a}	3.99 ± 0.02^{b}	36.65 ± 0.35^{a}	33.07 ± 0.16^{b}
T4	6.17 ± 0.03^{a}	5.58 ± 0.03^{b}	3.37 ± 0.02^{a}	3.16 ± 0.02^{b}	45.10 ± 0.37^{a}	35.49 ± 0.16^{b}
T5	8.82 ± 0.02^{a}	3.95 ± 0.02^{b}	2.86 ± 0.07^{a}	2.62 ± 0.07^{b}	65.09 ± 0.32^{a}	61.36 ± 0.16^{b}
T6	6.00 ± 0.02^{a}	2.59 ± 0.02^{b}	2.47 ± 0.02^{a}	2.53 ± 0.03^{b}	71.36 ± 0.41^{a}	68.25 ± 0.16^{b}
T7	3.82 ± 0.02^{a}	3.25 ± 0.12^{b}	2.30 ± 0.08^{a}	2.35 ± 0.14^{a}	82.29 ± 0.34^{a}	84.21 ± 0.15^{a}

Values within the group with different superscripts differ significantly ($p < 0.05$)

T1: 1h before surgery, T2: 15 minutes after recovery from anaesthesia, T3: 4h after surgery, T4: 8h after surgery, T5: 24h after surgery, T6: 48h after surgery, T7: 7th day of surgery

Hancock (2005) reported higher University of Melbourne Pain Scale (UMPS) scores at all the observation intervals postoperatively in the animals undergoing ovariohysterectomy via traditional method in comparison to those sterilized by harmonic scalpel-assisted laparoscopy.

A significantly ($p<0.05$) lower AST values were recorded for LapOVE group at 15 minutes, 4h, 8h, 24h, 48h and on 7th day of surgery post-operative stages in comparison to the values recorded for OVE group (Table 1). This could be due to excess muscle trauma encountered in traditional ovariectomy in comparison to laparoscopic ovariectomy (Kaneko, 1980). Rangnath and Kumar (2007) also reported significant elevation ($p\leq0.05$) in the mean AST concentration in animals that were sterilized by left flank method of ovariohysterectomy in comparison to those sterilized by laparoscopic method of ovariohysterectomy between 48 to 72h post-operatively. Kandpal (2013) reported mean increase of plasma AST activity till 48 hrs postoperatively in both the groups undergoing ovariohysterectomy of which animals sterilized by laparoscopic method revealed little higher activity at 6h and 24 hrs.

Creatine kinase (CK) recorded for LapOVE group were significantly ($p<0.05$) lower in comparison to the values recorded for OVE group during the entire observation period. The values recorded at 8h, 24h and 48h were above the normal range in both the group (Table 1). In dogs, CK is mostly present in skeletal muscles, myocardium, brain and intestine and has been used as a marker of skeletal muscle damage. The specificity of CK measurement for the diagnosis of muscle disease is high but its sensitivity is low 0.83 and 0.32, respectively (Aktas *et al.*,1993). Although the conditions of anaesthesia and type of surgery may vary, CK activity is increased during and after surgery. Hancock *et al.* (2005) reported significant increase in mean serum CPK values from preoperative values, however there were no significant differences ($p=0.77$) in CPK values in animals undergoing ovariohysterectomy via Harmonic Scalpel-assisted laparoscopy and traditional celiotomy surgical groups at any time. Creatine phosphokinase was elevated above normal limits at 6h, 12h and 24h postoperatively in both groups.

Zapryanova *et al.* (2013) recorded creatine kinase activity in dogs with experimentally induced acute inflammation and concluded that in the experimental group, the plasma concentrations of the CK-activity were increased at the 48h (97.48±6.92 U/L) and remained significantly higher ($p<0.05$) at the 72h (97.43±2.93 U/L) compared to the control group (77.08±5.27 U/L).

C-reactive protein is the fastest reacting canine acute-phase protein, which increases in response to infection and tissue injury (Conner *et al.*, 1988). C-reactive protein (CRP) is a major acute phase protein showing increased serum concentrations in dogs with systemic inflammation following surgery, trauma, infections, or neoplasia. Significantly lower ($p<0.05$) CRP concentration were recorded in LapOVE group at 15 minutes, 4h, 8h, 24h, 48h and on 7th day postoperative stages in comparison to the values recorded for OVE group (Table 2). Rangnath *et al.* (2007) reported significantly higher CRP concentrations at 24h to 48h after operationin animals undergoing left flank ovariohysterectomy in comparison to those sterilized by laparoscopic method of ovariohysterectomy.

Assay of cortisol concentration has been used as an indicator of stress and pain in dogs (Hansen *et al.*, 1997; Malm *et al.*, 2004., Devitt *et al.*, 2005; Hancock *et al.*, 2005). Significantly lower plasma cortisol concentration were recorded in LapOVE group at 15 minutes, 4h, 24h

and 48 h postoperative stages in comparison to the values recorded in OVE group. Rangnath and Kumar (2007) reported significantly higher serum cortisol concentrations (24h to 48h after operation) in bitches undergoing left flank ovariohysterectomy in comparison to bitches that were sterilized by laparoscopic method of ovariohysterectomy. Devitt *et al.* (2005) also reported significant rise in the serum cortisol level for longer period in bitches sterilized by open method of ovariohysterectomy in comparison to those sterilized by laparoscopic method. Fox *et al.* (1994) and Smith *et al.* (1999) reported significant increase of plasma cortisol level in response to surgical stress in dog and cat after ovariohysterectomy.

A significantly ($p<0.05$) lower plasma glucose concentration were recorded for LapOVE group at 15 minutes, 4h, 8h, 24h and 48h post-operative stage in comparison to the values recorded for OVE group (Table 2). This could be due to more pain and stress encountered during open method. Laiju *et al.* (2011) also reported significant increase in blood glucose level after conventional open ovariohysterectomy method. Devitt *et al.* (2005) also observed significant increase in blood glucose concentration at 1, 2, 4 and 6 h following ovariohysterectomy, and at one hour following laparoscopic ovariohysterectomy in bitches. Rangnath *et al.* (2007) reported significantly higher blood glucose level in bitches undergoing left flank ovariohysterectomy in comparison to bitches that were sterilized by laparoscopic method of ovariohysterectomy during immediate post-operative period.

CONCLUSION

Based on the findings of present study, it can be concluded that laparoscopic ovariectomy is less stressful, less traumatic and less painfull in comparison to open ovariectomy and can be used as an alternate method to the open method with early recovery.

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