GASLESS LAPAROSCOPIC OVARIOHYSTERECTOMY AND COMPARISON OF SERUM LEVELS OF INTERLEUKIN-6 AND C-REACTIVE PROTEIN AFTER LOHE AND TRADITIONAL LAPAROSCOPIC OVARIOHYSTERECTOMY IN BITCHES

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Received: October 25, 2012 Accepted: February 13, 2013

Abstract

Ten mixed-breed female dogs were used in the study. Abdominal wall lifting was performed with a sterilised cotton strip. Four portal sites were used to complete gasless laparoscopic ovariohysterectomy (GLOHE) procedures. The proper and suspensor ligament, ovarian pedicle, and broad ligament of the uterus, uterine artery and vein were coagulated using bipolar electrocoagulation. After the uterine body was coagulated and cut, the end of the cervix was ligated with a loop suture. Blood samples were obtained before the surgery, immediately after the procedure, and on 1, 3, and 5 d postoperatively for the determination of interleukin-6 (IL-6) and C-reactive protein (CRP). No significant surgical complications occurred. After gasless laparoscopic ovariohysterectomy, a significantly higher serum IL-6 level was found immediately after the surgery and 1 d postoperatively when compared with the level observed after traditional laparoscopic ovariohysterectomy (LOHE). However, no significant differences were observed in CRP level between GLOHE and LOHE. GLOHE proved to be a safe and feasible procedure in bitches.

Key words: bitches, laparoscopy, ovariohysterectomy, IL-6, C-reactive protein.

Ovariohysterectomy (OHE), the most common surgical procedure in veterinary practice, provides a method for population control and a treatment of pyometra in dogs (4, 33) Clinically, multiple surgical techniques have been described for OHE in bitches, including open, laparoscopic, and laparoscopic-assisted procedures (2, 8, 9, 11, 14, 17). Compared with open OHE, laparoscopic ovariohysterectomy (LOHE) has the advantages of superior visualisation, smaller abdominal incisions, less pain, and shorter convalescence (10, 12, 15, 34). Nowadays, more studies on LOHE are performed in veterinary medicine.

Laparoscopy requires establishing the pneumoperitoneum to provide visualisation and operating space. Creation of the pneumoperitoneum is the first and most critical step of a laparoscopic procedure (22). Most commonly used gas for insufflation is CO₂. The application of CO₂ can cause several pathophysiologic changes, such as acid-base disturbances (hypercarbia) (13, 18), increased pulmonary pressure, bradarrhythmis, asystole (19, 20), reduction of splanchnic perfusion (intestinal ischemia) (18-20), and growth and metastases of tumours (3, 16, 28).

C-reactive protein (CRP) is a liver-derived acute phase protein; whereas, interleukin-6 (IL-6) is thought to play a crucial role in the pathogenesis of surgical trauma. Thus, the serum CRP and IL-6 values can be used to monitor the severity of trauma (29, 30).

To reduced side effects of CO₂, gasless laparoscopic technique has been applied in human medicine (21, 32). The purpose of the study was to describe the application of gasless laparoscopic technique in ovariohysterectomy (GLOHE) in bitches and compare the serum levels of IL-6 and CRP in GLOHE and LOHE treated animals.

Material and Methods

Ten healthy mixed-breed bitches, aged 12 to 24 months, weighing 10 to 23 kg, were used. The animals were randomly assigned to GLOHE group (n=5, one parous bitch) and LOHE group (n=5, two parous bitch). All bitches were determined to be healthy on the basis of physical examination and complete blood count (CBC). Feed was withheld for 12 h, and water for 6 h. The bitches were given medetomidine (40 μg/kg, Hisunny Chemical Co., Ltd., China), and butorphanol (0.2 mg/kg, Jiangsu Hengrui Medicine Co., Ltd., China) and midazolam (0.5 mg/kg, Yichang Humanwell Pharmaceutical Co., Ltd., China) mixture intramuscularly, 20 min later.

Blood samples were obtained before the surgery,
immediately after the procedure, and on days 1, 3, and 5 postoperatively for the measurement of serum CRP and IL-6 levels by ELISA. All animal procedures were approved by the Laboratory Animal Care and Use Committee of Heilongjiang Province.

**GLOHE.** Four portal sites (Fig. 1) were used: portal A or B for a laparoscope (0°, Olympus, Germany) and portals C and D for surgical instruments. Portal A was 5 cm to the left side of the umbilicus. Portal B was 5 cm to the right side of the umbilicus. Portals A and B were symmetrical in respect to the umbilicus. Portal C was 5 cm cranial to the pubis and 5 cm to the left side of the linea alba. Portal D was located at the same level as portal C, but on the right side of the abdomen.

![Fig. 1. Localisation of portal sites](image)

Four incisions were made at four portal sites. The end of a sterilised cotton strip (100 cm long, 1 cm width) was introduced into abdomen at portal C and pulled out using a haemostat at portal D. Then, the cotton strip was pulled out at portal B and portal A (Fig. 1). These procedures were completed by laparoscopic guidance. The four ends of cotton strips were clamped and suspended to make abdominal wall lifting.

A 10/11 mm trocar-cannula unit was inserted through the abdominal wall at portals A and B, and a 5/5.5 mm trocar-cannula unit was inserted at portals C and D after abdominal wall lifting was completed. Laparoscope was inserted into abdomen at portal A, atraumatic grasping forceps at portal B, bipolar electrocoagulation (5 mm diameter, 330 mm long; Olympus, Germany) at portal C when the right uterine horn and ovary were isolated. Portals B, A, and D were used similarly when the left uterine horn and ovary were isolated. The proper and suspensory ligament, ovarian pedicle, and broad ligament of the uterus, uterine artery and vein were coagulated using bipolar electrocoagulation and then transected using laparoscopic scissors. The uterus was coagulated and cut similarly cranial to the cervix. The end of the cervix was ligated with a modified Roeder knot outside the peritoneal, then pushed into abdomen and tied by laparoscopic knot pusher (5 mm diameter, 320 mm long; Optclla Medical instrument Co., Ltd, China). The detached uterus and ovaries were removed using a retrieval bag. A 2-0 polyglycolic acid suture was used to close the portal incisions in a simple interrupted pattern.

**LOHE.** Three portal sites were used: portal A for the laparoscope and portals B and C for surgical instruments. Portal A was located at the umbilicus. Portal B was 5 cm cranial to the pubis and 5 cm to the left side of the linea alba. Portal C was located at the same level as portal B, but on the right side of the abdomen. A 10/11 mm trocar-cannula unit was inserted through the abdominal wall at portals A and B, and a 5/5.5 mm trocar-cannula unit was inserted at portal C.

Pneumoperitoneum was established with an automatic high-flow insufflator to an intra-abdominal pressure of 8 mm Hg using carbon dioxide gas. Laparoscopic procedures were the same as GLOHE.

**Postoperative care.** Systemic antibiotics (ampicillin, 20 mg/kg, i.m., q 8 h) were administered for 3 d. Dog’s attitude and appetite were observed. Body temperature and heart, and respiratory rates were monitored until their values had returned to normal. The CBC (once daily) was measured 5 d postoperatively.

**Repeat laparoscopy.** On 10 d postoperatively, laparoscopic examination was performed to re-evaluate the entire abdominal cavity. Portal was located 10 cm cranially to the pubic on the linea alba.

**Statistical analysis.** Standard statistical methods were used for the analysis of all results. Statistical differences within each group were determined by one-way ANOVA. Paired-Samples T test was used to compare the two groups. Significant level was established as P<0.05. Statistical analysis of data was performed with computer software (SPSS, USA).

**Results**

In the study, LOHE and GLOHE were successfully completed in 10 dogs. No major surgical complications occurred in either group. Minor haemorrhage was observed during surgical procedures but it was insignificant. LOHE resulted in only two 11 mm and one 5.5 mm incisions in the body wall, and GLOHE was done by two 11 mm and two 5.5 mm incisions in the wall. During surgical procedures, exposure of visualisation in LOHE group was better than in GLOHE group. The median length of surgery was 55 min (ranged 45-75 min) for LOHE and 60 min (ranged 50-82 min) for GLOHE.

During second-look laparoscopy on 10th day postoperatively, no abdominal abnormalities were noticed in LOHE; however, a focal fibrous omentum to the abdominal wall was attached in GLOHE.

Serum IL-6 levels increased significantly just after the procedure, as well as on day 1 and 3 post surgery in both GLOHE and LOHE (P<0.05), however, statistically
significant differences in serum IL-6 levels were noted between GLOHE and LOHE immediately after the procedure and 1 d postoperatively (P<0.05) (Fig. 2).

![Graph showing changes in serum C-reactive protein (CRP) and IL-6 levels](image)

**Fig. 2.** Changes in interleukin-6 (IL-6) in gasless laparoscopic ovariohysterectomy (GLOHE) and traditional laparoscopic ovariohysterectomy (LOHE) groups. * P<0.05 compared to LOHE, # P<0.05 compared to preoperative serum levels.

**Fig. 3.** Changes in C-reactive protein (CRP) in gasless laparoscopic ovariohysterectomy (GLOHE) and traditional laparoscopic ovariohysterectomy (LOHE) groups. * P<0.05 compared LOHE, # P<0.05 compared to preoperative serum levels.

Serum CRP levels increased significantly immediately after the procedure and 1 d post operation in GLOHE and LOHE, and also on 3rd day postoperatively in LOHE (P<0.05). A comparison between GLOHE and LOHE showed no significantly differences. (Fig. 3)

**Discussion**

Intra-abdominal pressure of 8 mm Hg that was used to satisfactorily separate the abdominal wall from visceral structures, providing enough operation space and excellent observation to view the bilateral ovary and pedicle under laparoscopic guidance in LOHE. Abdominal wall lifting techniques rapidly developed, since gasless laparoscopy has been firstly applied in cholecystectomy by Nagai in 1991 (1, 5, 6, 24, 27). These techniques provided a good visualisation for surgical procedures. In the study, abdominal wall lifting was performed using four portals with a sterilised cotton strip. Surgical procedures were successfully completed by this simple method, but exposure of visualisation in GLOHE was worse than in LOHE. After suspending the ends of cotton strip, vertical diameter increased and transverse diameter decreased. Exposed space of the ovarian pedicle also decreased, but an excellent exposure of the uterine artery and vein was observed. Therefore, abdominal wall lifting technique should be further improved. During the second-look laparoscopy, a low frequency of abdominal adhesion was observed in GLOHE; however, the adhesion was not associated with any clinical problems.

The treatment of the ovarian pedicle and uterine artery and vein is a key to a successful ovariohysterectomy. Clips, endoloop, monopolar electrocaigation, bipolar electrocoagulation, neodymium: yttrium aluminum garnet surgical laser, and harmonic scalpel were used to solve such problems during laparoscopy (9, 11, 12, 14, 17, 34). The bipolar cautery method to seal vessels was used. This method produced a good effect. Minor haemorrhage was observed during surgical procedures but it was insignificant. Production of smoke was observed, especially in obese dogs and parous dogs, but this could be aspirated with a suction irrigation tube.

IL-6 production and activation by monocytes, macrophages, and endothelial cells is an early response to surgical trauma, and an increased level of this cytokine was observed in the circulation after the used procedures. It has been shown that IL-6 serum level is almost proportional to the severity of the surgical trauma (23). In the above study, a significant increase in serum IL-6 after surgical procedure of GLOHE and LOHE was found. The increase indicates a significant trauma after GLOHE and LOHE, nevertheless, a lesser trauma was observed in LOHE comparing to GLOHE. The cotton strip for abdominal wall lifting scraped portal incision, which might have been an important stress to the body, elevating the serum IL-6 level and creating the abdominal adhesion in GLOHE.

CRP is a key marker of acute-phase proteins, and its increased concentration represents a consistent inflammatory response to trauma and infections. IL-6 precedes and directs the hepatic release of CRP (31). Serum levels of CRP were increased significantly in both groups after surgery. Comparing to LOHE, levels of CRP was higher in GLOHE, but no statistical differences in CRP were found in both groups. Serum CRP levels did not reflect trauma difference between GLOHE and LOHE.

**Acknowledgments.** This work was supported by the Educational Commission of Heilongjiang Province of China No. 12511033 and the Doctoral Research Foundation of Northeast Agricultural University No. 2012RCB23.
References


