

## Comparison of Two Techniques on Titanium Clip Application for Ovariohysterectomy in Bitches

Jin-Seok Park, Hyun-Gu Kang<sup>1</sup> and Ill-Hwa Kim

*Department of Veterinary Obstetrics and Theriogenology, College of Veterinary Medicine, Chungbuk National University, Cheongju 361-763, Korea*

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**Abstract :** The objective of this study was to assess the complications of titanium clip application after ovariohysterectomy (OHE) and find the proper surgical technique to minimize the complications by comparing the ligation method of ovarian pedicle. Six female beagle dogs were used for this study and they were divided randomly into two groups by ligation techniques: ligation and non-ligation of the suspensory ligament of ovary including ovarian pedicle. To evaluate the difference between two techniques, the location and embedded pattern of applied clips in the abdominal cavity were identified through radiographic tests and autopsy. Hematology tests were also performed to check the existence of inflammation by applied clips and all tests were carried out monthly for 1 year. There were few changes in location of clips of both groups on X-ray views without any migration. Almost all values in hematology of two groups were within the normal range and there were no inflammatory symptoms. All of applied clips in two groups were found near the kidney, but the embedded forms of them showed slight differences. The clips ligating suspensory ligament and ovarian pedicle were located in deep place and thickly surrounded with thin layers of peritoneum. But the clips ligating only ovarian pedicle were found with ease on surface of layer and located in lower place compared with the clips ligating suspensory ligament. These results indicate that the suspensory ligament holds applied clips and minimizes the movement of them. Therefore, ligation of suspensory ligament would be the preferred technique in titanium clip application for OHE.

**Key words :** ovariohysterectomy, titanium clip, ovarian pedicle, suspensory ligament.

### Introduction

Ovariohysterectomy (OHE) is a surgical procedure widely employed in veterinary practice. The most common reason to perform OHE is to prevent estrus and unwanted offspring and it is also indicated in cases of pyometra, uterine tumors, or other pathologies (1,4). Many technical variations of OHE have been described, including flank and laparoscopic approaches and the use of stapling equipment, ultrasonic scalpel, vessel sealing devices, or Miller's knots (1,4,10). But OHE is traditionally performed through a small ventral midline incision (5). Several techniques have been described for ligating the ovarian and uterine pedicles in traditional OHE. Among them, double ligations, utilizing an encircling and a transfixing ligature are recommended on all ovarian pedicles in adult dogs (5).

Recently, instead of an encircling and a transfixing ligature, the metal clip is used for ligation. The clip application comes into wide use because it is an easier operation to perform and results in a significant reduction in the operative time

(10). The metal clip must be highly biocompatible and must provide adequate permanent closing force. The titanium used for ligature clips is a biocompatible material with adequate strength and resistance to corrosion (9). The reliability of the clip material has been tested and approved by International Organization for Standardization (6).

This clip application has been used for various surgical techniques in human medical fields. Also it has been studied about the complications of each surgery used clip application. Among them, some complications of Filshie clip application have been reported in the medical literature of obstetrics and gynecology (3,7,8,11,13). Filshie clip is a small, hinged titanium clip lined with a soft silastic rubber and it is used for female sterilization applying tubal occlusion (12). The complications of Filshie clip include pelvic pain and intraperitoneal migration into the bladder, appendix and vagina (3,7,8,11,13). But, in case of OHE in the dog and cat, although the titanium clip application is currently used widely, there are comparatively few reports in the literature regarding the complication.

Therefore, the purpose of this study was to evaluate the complication of titanium clip application after OHE and find the proper surgical technique to minimize the complication by comparing two ligation methods: ligation and non-ligation of the suspensory ligament of ovary including ovarian pedicle.

<sup>1</sup>Corresponding author.  
E-mail : kang6467@chungbuk.ac.kr

## Materials and Methods

### Experimental animals

Six female beagle dogs (1 year old) were used for this study. The animals were obtained from Samtako (Beijing Marshall Biotechnology Co, Ltd, Beijing, China) and housed separately in stainless net cage. Nutritional formula dog food was fed to the animals once a day, water was fed freely, and they took anthelmintics (ivermectin and pyrantel pamoate) every month. Because it took 1 year to estimate the results of surgery, they were housed in conditions mentioned above for 1 year. They were divided randomly into two groups of three beagle dogs for OHE.

### Surgical procedures

All procedures were performed entirely applying general OHE surgical techniques (1,5) except using the suture of ovarian pedicle. Before the surgery, it was ensured that each dog has been fasted since the previous day. After induction of anesthesia with Zoletil 50<sup>®</sup> (2.5 mg/kg, Virbac, Carros, FRA), isoflurane was used for maintaining through inhalation. The laparotomy was proceeded along the linea alba in accordance with general surgery procedures. After identifying the left uterine horn and ovary in the bottom of the surgical field, the medium-large titanium clips (HORIZON<sup>™</sup>, Weck Closure Systems, North Carolina, USA) were applied for ligation of ovarian pedicle using clip applier. The opposite ovary was also performed as just described. When the titanium clips were applied to both ovarian pedicles, one of two groups was ligated including the suspensory ligament and ovarian pedicle, but the other one was ligated just including ovarian pedicle to compare the location of clips in the abdominal cavity. After sectioning the ovarian pedicle and broad ligament, the cervix was double ligated with suture not using clips and then the uterine body was transected. The Portal closure and recovery from anesthesia were subsequently proceeded and each dog received fluximine (1 mg/kg SC) every 12 hours for 3 days and enrofloxacin (2.5 mg/kg SC) for 1 week.

### Radiographic tests

Abdominal X-ray tests (10 mAs, 80 kVp, BLD-150RK, LISTEM, Wonju, Korea) were performed monthly for 1 year to identify the location of clips in ventrodorsal and lateral position. Before the X-ray tests, each dog was fasted for 24 hours to reduce the change of clip location by dilation of the

stomach in the position view.

### Blood tests

Hematology tests were performed to check the existence of inflammation by clips on the same day of X-ray tests. Blood samples were collected from the jugular vein by using 23 gauge syringe and gathered into the EDTA-3K bottles (Sewon Medical, Busan, Korea). Hematology were analyzed within 20 minutes after collection using a blood analyzer (CELL-DYN<sup>®</sup> 3700, ABBOTT, Illinois, USA) and the following parameters were evaluated: white blood cell count, differential leukocyte count, red blood cell (RBC) count, hemoglobin concentration, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, RBC distribution width, platelet count and mean platelet volume.

### Autopsy

The location and embedded pattern of applied clips in the abdominal cavity was identified through autopsy after 1 year post-OHE. Autopsy was performed following euthanasia using Zoletil 50<sup>®</sup> (2.5 mg/kg, Virbac, Carros, FRA) and KCl (Daihan Pharm Co, Ltd, Seoul, Korea). After euthanasia, left and right flanks of each dog were incised near the kidney and then peritoneum was opened. We grossly observed the location and embedded pattern of applied clips.

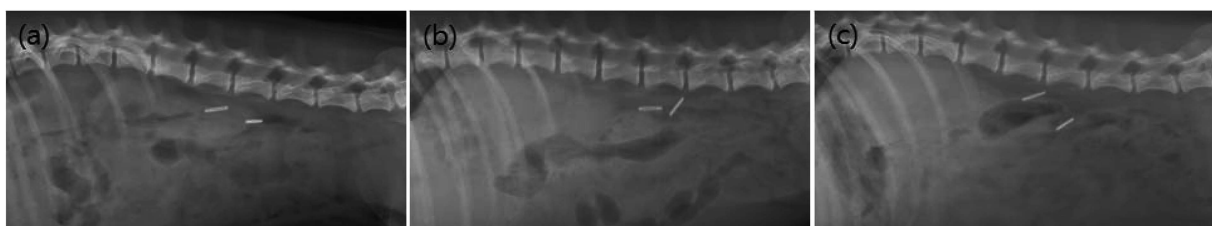
## Results

### Surgical complications

All of six beagle dogs recovered uneventfully from OHE surgery. A small quantity of hemorrhage in the surgical field from the ovarian pedicle occurred in all dogs. But the amount of blood loss was considered clinically inconsequential. All dogs remained hemodynamically stable and no other additional treatments such as blood transfusions were administered.

### Radiographic view

The applied clips on the lateral and ventrodorsal X-ray view were observed near the kidney. As time passed since clip application OHE, there was few change in location of clips of all six cases on both X-ray views without any movement to inferior or caudal region in the peritoneal cavity (Figs 1 and 2). In addition, there was no radiographic abnormality related with clip induced OHE complications.



**Fig 1.** Lateral X-ray views of abdominal cavity after clip application for ovariohysterectomy; (a) 30 minutes later, (b) 6 months later and (c) 1 year later.

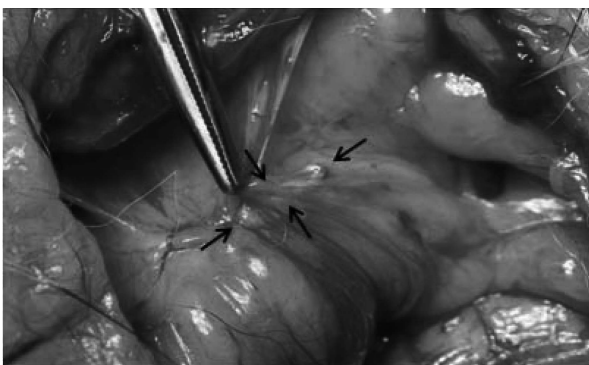


**Fig 2.** Ventrodorsal X-ray views of abdominal cavity after clip application for ovariectomy; (a) 30 minutes later, (b) 6 months later and (c) 1 year later.

**Table 1.** Hematology tests results of 6 female beagle dogs after 1 month, 6 months and 12 months post-ovariohysterectomy

Parameter	Value (mean $\pm$ SD)			Unit	Normal Range
	1 mo	6 mo	12 mo		
White blood cell	10.87 $\pm$ 2.29	7.64 $\pm$ 1.23	9.13 $\pm$ 0.51	K/ $\mu$ L	6.00 - 17.00
Neutrophil	7.91 $\pm$ 1.50	6.19 $\pm$ 1.01	5.16 $\pm$ 0.35	K/ $\mu$ L	3.62 - 11.32
Lymphocyte	1.54 $\pm$ 0.83	2.64 $\pm$ 0.92	2.87 $\pm$ 0.62	K/ $\mu$ L	0.83 - 4.69
Monocyte	0.67 $\pm$ 0.25	0.68 $\pm$ 0.31	0.47 $\pm$ 0.09	K/ $\mu$ L	0.14 - 1.97
Eosinophil	0.67 $\pm$ 0.45	0.37 $\pm$ 0.34	0.37 $\pm$ 0.11	K/ $\mu$ L	0.04 - 1.56
Basophil	0.05 $\pm$ 0.04	0.03 $\pm$ 0.02	0.10 $\pm$ 0.02	K/ $\mu$ L	0.00 - 0.12
Red blood cell	6.75 $\pm$ 0.53	6.53 $\pm$ 0.46	6.34 $\pm$ 0.68	M/ $\mu$ L	5.10 - 8.50
Hemoglobin	12.3 $\pm$ 1.22	15.5 $\pm$ 0.81	15.4 $\pm$ 1.48	g/dL	11.0 - 19.0
Hematocrit	42.4 $\pm$ 2.67	44.2 $\pm$ 2.11	41.1 $\pm$ 3.18	%	36.0 - 56.0
MCV	63.0 $\pm$ 2.40	67.9 $\pm$ 2.48	65.1 $\pm$ 2.93	fL	60.0 - 74.0
MCH	18.3 $\pm$ 1.47	23.8 $\pm$ 0.79	24.4 $\pm$ 0.92	pg	19.5 - 24.5
MCHC	29.0 $\pm$ 2.04	25.8 $\pm$ 4.84	37.4 $\pm$ 1.15	g/dL	30.0 - 38.0
RDW	16.1 $\pm$ 0.62	17.1 $\pm$ 1.36	12.3 $\pm$ 0.40	%	11.5 - 15.9
Platelet	331 $\pm$ 57.3	268 $\pm$ 49.4	262 $\pm$ 65.8	K/ $\mu$ L	117 - 460
Mean platelet volume	13.0 $\pm$ 1.08	10.8 $\pm$ 1.68	7.5 $\pm$ 0.43	fL	7.3 - 11.2

SD, standard deviation; mo, month; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; RDW, red blood cell distribution width.



**Fig 3.** Applied titanium clip (arrows) ligating suspensory ligament and ovarian pedicle after 1 year post-ovariohysterectomy.

### Hematology

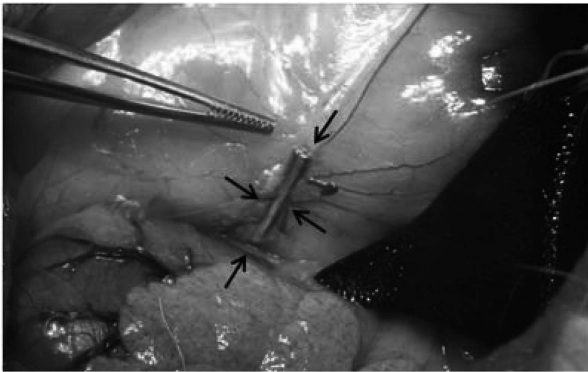
The hematology tests results after OHE are shown in Table 1. Almost all values in hematology were within the normal range and there were no inflammatory symptoms.

### Assessment of applied clips

All of applied clips were found near the kidney without any dislocation. But the embedded form of applied clips showed slight difference between two groups. In case of the clips ligating suspensory ligament and ovarian pedicle, they were located in deep place and thickly surrounded with thin layers of peritoneum (Fig 3). But the clips ligating only ovarian pedicle were found with ease on surface of layer and located in lower place compared with the clips ligating suspensory ligament and ovarian pedicle (Fig 4). The ovarian artery and vein, ligated with clips, were almost degenerated and there were no hemorrhage evidences.

### Discussion

The titanium clips are widely used for ligation of ovarian pedicle in OHE, because the titanium is more biocompatible and its mechanical properties are more reliable without pathogenomic complications (6,9). They are also used for sterilization of women in human medicine. The Filshie clip steriliza-



**Fig 4.** Applied titanium clip (arrows) ligating only ovarian pedicle after 1 year post-ovariohysterectomy.

tion is an effective and immediate method of female sterilization, which is made by titanium and silastic rubber (12). The titanium clips effect ligation by being locked across the breadth of ovarian pedicle. Avascular necrosis occurs at the site of clip application with ovarian pedicle leaving healed and occluded stumps (7). Generally the titanium clip remains attached at the site of ligation, eventually becoming covered by a thin layer of peritoneum. The function of clip is applied to the sterilization of women identically. However, occasionally it was reported in human medical fields that they were dislodged and settled elsewhere within the peritoneal cavity or extruded through genital organs (3,7,8,11,13). Therefore, it is necessary to verify the complications of applied clips after OHE such as dislodgement, inflammation or extrusion. Also, it is important to make sure that how they are settled and embedded in the peritoneal cavity.

In this one year study, the applied clips of six female beagle dogs were not dislodged in the peritoneal cavity and they were observed near the kidney (Figs 1 and 2). Some of clips showed that their location moved just a little bit lower on the lateral X-ray view compared with the previous view (Fig 1). It is thought that those changes are due to body weight gain and accumulation of fat in the retroperitoneal space.

If the clips move to inferior or caudal region in the peritoneal cavity, they can occur inflammation and abdominal pain by causing damage to the abdominal organs. These clinical sings were reported in some medical journals of human obstetrics and gynecology (3,7,8,11,13). Controlled studies performed in human have shown that Filshie clips are capable of inducing a low grade inflammation by the adhesion formation around dislodged Filshie clips (2). The dislodged clip can be migrated transperitoneally and extruded through bladder, rectum and vagina by a low grade inflammation. Similarly, it is very necessary to check the inflammation or abdominal pain after OHE surgery applying titanium clips in the female dogs. In blood tests of six female dogs after OHE, almost all of hematology values were within normal ranges (Table 1) and they had no clinical signs related with inflammation. These results can be associated with no migration of titanium clips and confirm through the radiographic results described above

(Figs 1 and 2).

The slight differences between two groups were found in the location and embedded form. The clips ligating suspensory ligament and ovarian pedicle were thickly surrounded with thin layers of peritoneum and they were located in deep place behind the kidney (Fig 3). However, the clips ligating only ovarian pedicle were surrounded with layers very thinly and they were found under the kidney (Fig 4). The difference between two results means that the suspensory ligament holds ligation clips and minimizes the movement of them. Also, these results imply that ligation method of suspensory ligament is safer than that of non-ligation of suspensory ligament in that it can reduce complications by migration.

The period of one year and relatively small number of patients in this study means that no significant difference in surgery method could be established between ligation of suspensory ligament and non-ligation of suspensory ligament in the clip application for OHE. Nevertheless, our experience would indicate that ligation of suspensory ligament is the preferred technique because of no migration of clip and minimization of complication, and the titanium clip application is very reliable for OHE surgery without specific complications.

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## 개의 난소자궁적출술에 있어서 티타늄클립의 두 가지 적용방법에 대한 비교

박진석 · 강현구<sup>1</sup> · 김일화

충북대학교 수의과대학

**요 약** : 본 연구에서는 난소자궁적출술에 있어서 티타늄(titanium) 클립 적용 시 발생할 수 있는 합병증을 평가하고, 난소혈관과 난소결이인대의 두 가지 결찰법을 비교하여 합병증을 최소화 할 수 있는 수술법을 찾고자 하였다. 실험을 위해 6마리의 암컷 비글견이 사용되었으며, 세 마리씩 두 그룹으로 나누어 한 그룹은 난소결이인대와 난소혈관을 함께 결찰하였고, 나머지 한 그룹은 난소결이인대를 결찰하지 않고 난소혈관만 결찰하였다. 두 수술법을 비교하기 위해서 매달 1회씩 1년 동안 복강 X-ray 검사를 통해 적용된 클립의 위치 변화를 확인하였고, 적용된 클립에 의한 염증반응 유무는 혈액검사를 통해 확인하였다. X-ray 검사 상에서 두 그룹의 클립 위치는 큰 변화를 보이지 않았으며, 혈액 검사에서도 거의 모든 수치들이 정상범위에 있었고, 염증과 관련된 임상증상 또한 관찰되지 않았다. 부검을 통한 두 그룹의 클립 포매 형태를 관찰한 결과 난소결이인대를 결찰한 클립은 복강의 얇은 막으로 두껍게 싸여서 신장 후면에 위치하고 있는 것을 확인하였고, 난소결이인대를 결찰하지 않은 클립은 얇게 둘러싸인 막 표면에 위치하여 신장의 아래쪽에서 관찰되었다. 이러한 결과는 난소결이인대가 클립의 움직임을 최소화 할 수 있도록 지지해주는 역할을 하는 것을 의미하며, 따라서 티타늄 클립을 적용하는 난소자궁적출술에서 난소결이인대를 함께 결찰하는 방법이 합병증 예방에 있어서 보다 적합한 방법으로 생각된다.

**주요어** : 난소자궁적출술, titanium clip, 난소혈관, 난소결이인대