# In situ coagulation and transection of the ovarian pedicle: An alternative to laparoscopic ovariectomy in juvenile horses

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## Abstract

The feasibility of leaving the ovaries within the peritoneal cavity after laparoscopic coagulation and transection of the ovarian pedicle was assessed in the juvenile horse. Elective ovariectomy was performed on 10 quarter horses, aged 4 to 5 mo, with the fillies in a Trendelenburg position. The mesovarium was isolated, and multiple coagulation and transection cycles were performed until all ovarian attachments had been severed. The ovaries were dropped within the abdomen, and hemostasis of the transected mesovarium was evaluated before closure. The mean surgical time was 33 min (range, 23 to 48 min). Ten weeks after surgery the fillies were humanely euthanized. At postmortem examination, the ovary location within the abdomen was noted. In 1 horse, there was an abdominal adhesion; viscera had been punctured during insufflation. Of the 20 ovaries, 4 were free-floating within the abdominal cavity; the other 16 were enveloped in the free portion of the greater omentum in the cranioventral abdomen. Histologic examination of the ovaries was performed to assess follicle cell viability. In both the free-floating and the attached ovaries, the deep blood vessels and all examined follicular structures were necrotic and partially mineralized. Laparoscopic electrosurgical transection of the ovarian pedicle without removal of the ovaries should be considered an alternative to other ovariectomy techniques that may be performed in young female horses.

## *Résumé*

*La faisabilité de laisser les ovaires à l'intérieur de la cavité péritonéale après coagulation et trans-section du pédicule ovarien par laparoscopie a été évaluée chez des chevaux juvéniles. Une ovariectomie élective a été effectuée chez 10 chevaux Quarter horse, âgés de 4 à 5 mois, avec les juments dans une position Trendelenburg. Le mesovarium a été isolé, et des cycles multiples de coagulation et de trans-section ont été effectués jusqu'à ce que les attaches ovariennes aient été coupées. Les ovaires ont été laissées libres dans l'abdomen, et l'hémostase du mes-ovarium trans-sectionnné a été évaluée avant fermeture. Le temps moyen de chirurgie était de 33 min (écart, 23 à 48 min). Dix semaines après la chirurgie, les juments ont été euthanasiées. Lors de l'examen post-mortem, la localisation intra-abdominale des ovaires a été notée. Chez 1 cheval il y avait une adhésion abdominale; les viscères avaient été perforés au moment de l'insufflation. Parmi les 20 ovaires, 4 étaient libres dans la cavité abdominale alors que les 16 autres étaient enveloppés dans la portion libre du grand omentum dans la partie crânio-ventrale de l'abdomen. L'examen histologique des follicules a été effectué afin d'évaluer la viabilité cellulaire des follicules. Autant chez les ovaires libres que ceux attachés, les vaisseaux sanguins profonds et toutes les structures folliculaires examinées étaient nécrotiques et partiellement minéralisées. La trans-section électrochirurgicale par laparoscopie du pédicule ovarien sans le retrait des ovaires devrait être considéré comme une alternative aux autres techniques d'ovariectomie pouvant être effectuées chez de jeunes juments.*

*(Traduit par Docteur Serge Messier)*

## Introduction

Bilateral ovariectomy is an elective procedure commonly used to prevent pregnancy, manipulate the estrus cycle, manage colic associated with estrus, and modify behaviour associated with the estrus cycle (1,2). In an extensive survey of horse owners in 1996, members of the American Association of Equine Practitioners reported that the estrus cycle has a negative impact on the performance of athletic females (3). Horse owners have reportedly discriminated against using intact mares for athletic endeavours because of undesirable behaviour during estrus (1).

Treatment options to minimize undesirable influences of estrus are limited to medical management with exogenous progestins or surgical removal of both ovaries. Progestins are expensive, daily administration is required when they are given orally, and there are health risks to the person administering the daily medication (4). In our opinion, high morbidity rates with traditional ovariectomy techniques have led to reluctance by owners to have the procedure

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performed. Surgical methods of ovariectomy include colpotomy, flank incisions, ventral incisions, and, more recently, laparoscopic techniques (5–12). Serious complications associated with colpotomy for ovary removal include exsanguination, shock, adhesions, abdominal pain, and peritonitis (1,6). Body wall incisions for ovary removal have been reported to require lengthy convalescence and to have incisional complications ranging from seroma formation to dehiscence (13). These complications have led to the development of less invasive laparoscopic techniques (8,11). Laparoscopic ovariectomy (ventral or flank) provides excellent visualization of the ovary and mesovarium and allows tensionless manipulation of the mesovarium during pedicle transection, visual assessment of hemostasis, smaller body wall incisions, and, in some cases, removal of both ovaries through the same operative site (8–12,14).

The numerous methods used for hemostasis of the mesovarium and the associated ovarian vessels have included ligature application (10), laser techniques (11), electrocoagulation (12,15), and use of the harmonic scalpel (16), stapling instruments (17), and vascular clips (18). Common problems are technical difficulty applying the hemostatic devices and the expense associated with consumables. Ligature slippage has also been associated with inadequate hemostasis (19). Compared with other methods, electrocoagulation has resulted in reliable hemostasis, is technically straightforward, and is inexpensive (12).

Laparoscopic surgery can be technically demanding for the operator. Operative time is directly related to the surgeon's experience. After transection of the 1st ovary, the surgeon usually needs to identify an area to store the ovary before removing the 2nd ovary to prevent loss of the pneumoperitoneum (8). If an ovary is inadvertently dropped during removal and cannot be observed with the laparoscope and grasped, the standard of practice is to remove the instrument cannula and extend the incision to allow manual retrieval of the dropped ovary. Extending the incision negates the advantage of minimal invasiveness associated with laparoscopy. The consequence of failing to remove the transected ovary is unknown.

It is common practice to leave amputated ovaries free in the abdomen of cattle undergoing ovariectomy via colpotomy, and no detrimental effects have been reported (20). If ovariectomies in the horse could be performed satisfactorily with laparoscopic coagulation and transection of the ovarian pedicles without removal of the organs, numerous advantages could be realized in addition to rendering the ovaries nonfunctional. Leaving the ovaries within the abdomen would improve on currently described laparoscopic techniques by reducing surgical time and technical demands on the surgeon, improving cosmesis, and minimizing the risk of incisional complications. The purpose of this project was to assess the feasibility of laparoscopic ovariectomy without removal of the ovaries from the abdomen of juvenile horses.

# Materials and methods

With approval of the University of Saskatchewan Animal Care and Use Committee, laparoscopic ovariectomy was performed on 10 healthy quarter horse fillies aged 4 to 5 mo and weighing 160 to 180 kg. The animals were housed together in a large paddock and had unlimited access to feed and water. Before surgery, feed was withheld for 24 to 36 h. All the fillies received a tetanus toxoid (Ayerst, Guelph, Ontario) booster intramuscularly before surgery. Procaine penicillin G (Ethacillin; Roger/STB, London, Ontario), 22 000 U/kg, was administered intramuscularly 1 h before surgery and once after surgery. The horses received flunixin meglumine (Banamine; Schering–Plough, Pointe Claire, Quebec), 0.5 mg/kg intravenously every 8 h, beginning 1 h before surgery and continuing for 48 h. During surgery, heart rate, direct arterial blood pressure, and respiratory rate were monitored, and electrocardiographic analysis was performed. Surgical time (defined as the time from initial insertion of the laparoscope to incision closure), intraoperative and postoperative complications, perioperative attitude, and postmortem and histologic findings were recorded.

Anesthesia was induced with a combination of xylazine hydrochloride (Rompun; Bayer, Etobicoke, Ontario), 1.0 mg/kg, ketamine hydrochloride (Vetalar; Vetrepharm, London, Ontario), 2.0 mg/kg, and diazepam (Sabex; Boucherville, Quebec), 0.02 mg/kg, administered intravenously. After endotracheal intubation, the fillies were placed in dorsal recumbency with their hindlegs tied to the surgical table. Anesthesia was maintained with halothane in oxygen, delivered through a rebreathing system. Ventilation was assisted with positive-pressure ventilation. All animals received butorphanol tartrate (Torbugesic; Ayerst), 0.05 mg/kg, administered intravenously, at the start of the surgical procedure. Surgical preparation included clipping the ventral abdomen from the xyphoid to the inguinal region, followed by aseptic surgical preparation.

## Surgical procedure

A 2-cm-long skin incision was made 1 to 2 cm caudal to the umbilicus at the midline. A 5-mm-long stab incision was made in the middle of the skin incision through the linea alba, and a teat cannula was passed through the peritoneum into the abdominal cavity. Correct placement was confirmed by the ability to feel abdominal viscera with the end of the teat cannula. The teat cannula was connected to a high-flow electronic insufflator (Karl Storz Veterinary Endoscopy, Goleta, California, USA), and the abdomen was distended with carbon dioxide to a pressure of 15 mm Hg. The teat cannula was then removed, and a guarded sharp pyramidal trocar sheathed in a cannula with a diameter of 11 mm (Dr Fritz Endoscopy and Video Systems, Tutlingen, Germany) was thrust firmly into the abdominal cavity in the same location. Care was taken to ensure that pressure applied to the trocar–cannula unit was perpendicular to the linea alba. The sharp trocar was removed and replaced by a 10 mm × 33 cm, 30° laparoscope (Karl Storz Veterinary Endoscopy) connected to a 300-W xenon light source (Karl Storz Veterinary Endoscopy) and laparoscopic camera (Linvatec Canada, Mississauga, Ontario). Insufflation was maintained at a pressure of 10 to 15 mm Hg during the procedure. The surgery table was inclined to roughly 30°, with the head downward (Trendelenburg's position), to displace the abdominal viscera cranially.

After the caudal abdomen was observed, 2 instrument portals were made, 15 to 20 cm caudal to the laparoscope portal and 5 to 10 cm to the right and left of the midline. Two incisions, 10 to 15 mm long, were made through the skin and the external abdominal oblique fascia, parallel to the linea. Instrument cannulas 10 mm in diameter were placed with blunt trocars under direct observation

with the laparoscope. The instrument cannulas were fitted with adapters (Karl Storz Veterinary Endoscopy) that reduced the diameter of the portals from 10 mm to 5 mm to accommodate the 5-mm bipolar electrosurgical forceps (Kleppinger bipolar forceps; Richard Wolf Medical Instruments, Rosemont, Illinois, USA) and the 10-mm laparoscopic scissors. Laparoscopic atraumatic Babcock grasping forceps were inserted through both working portals, and the ovaries or uterine horns were located in the caudal abdomen abaxial and caudal to the apex of the bladder. Large and small intestine was gently manipulated to locate the reproductive tract, then traction was applied to the ovaries to allow manipulation of the ovarian pedicles. Atraumatic forceps passed through the ipsilateral portal were used to grasp an ovary at the cranial pole. The proper ovarian ligament was extended between the ovary and the tip of the uterine horn. Proper positioning was accomplished by passing the ovary back and forth between 2 atraumatic grasping forceps until the mesovarium was easily accessible.

The ovary was manipulated with a Babcock forceps placed through the ipsilateral portal to ensure that adequate distance was maintained between the mesovarium and the viscera. A 5-mm bipolar electrosurgical forceps was inserted through the contralateral instrument portal and placed across the cranial aspect of the mesovarium. Electrical current (11.5 W) was applied until the grasped tissue bubbled and turned white, indicating adequate coagulation. The tissue was then transected with laparoscopic scissors. Multiple coagulation and transection cycles were required to completely transect the ovarian pedicle. After the ovarian artery was transected, it was specifically targeted for additional electrocoagulation. Once coagulation and transection of the entire mesovarium was complete, the ovary was dropped in the abdomen, and the tip of the uterine horn and mesovarium were elevated and observed for hemorrhage. The 2nd ovary was coagulated, transected, and dropped in a similar manner.

After completion of the surgical procedure, the pneumoperitoneum was relieved by opening the valves of the 3 cannulas and placing manual pressure on the ventral abdominal wall. The incisions in the abdominal wall were apposed with 1 polyglyconate cruciate suture (Maxon; Tyco Health Group Canada, St. Laurent, Quebec). The skin incisions were closed with 3-0 poliglecaprone sutures (Monocryl; J & J Medical Products Ethicon, Markham, Ontario) in a continuous subcuticular pattern. The horses were then returned to a horizontal position, allowed to recover from anesthesia, and gradually introduced to feed over the next 8 h.

After close observation for 48 h after surgery, the fillies were returned to a large outdoor paddock for twice-daily examination for the remainder of the study. They were introduced into an antibiotic pharmacokinetic trial 6 wk after surgery and euthanized with a barbiturate 10 wk after surgery.

### Postmortem and histologic examination

Postmortem examination concentrated on location and retrieval of the transected ovaries, the state of the mesovarium, and intra-abdominal adhesion formation. The ovaries were photographed, transected longitudinally and processed for histologic examination. The ovarian remnants were fixed in 10% neutral buffered formalin, sectioned, and stained with hematoxylin and eosin. All serial sections were completely assessed.

## Results

The surgical procedure was performed by a board-certified surgeon and a surgical resident. The procedure was not considered technically difficult by either surgeon, and no major operative complications were experienced. There was a trend towards shorter surgical times as experience increased. The mean surgical time was 33 min (range, 23 to 48 min).

In 1 filly, the teat cannula used for insufflation was inadvertently placed into an abdominal viscus, as indicated by the presence of greenish liquid in the hub of the cannula. The cannula was removed and replaced with a clean teat cannula. This filly was not elevated into the Trendelenburg position until laparoscopic evaluation of the abdomen was performed. The punctured viscus was not located, and the surgical procedure was completed as planned. The horse showed no signs of discomfort or colic after the ovariectomy. At necropsy, a fibrous adhesion 2 cm in diameter was noted, extending from the tip of the cecum to the omentum.

The bipolar electrosurgical system was easy to use and provided adequate coagulation of the mesovarium. The proper positioning of the ovary (outstretched proper ovarian ligament) with the grasping forceps was vital to efficient application of the electrosurgical forceps across the mesovarium. The sequential coagulation and transection was considered the rate-limiting step of the surgical procedure. Roughly 5 mm of tissue could be coagulated at 1 time; on average, 6 to 10 coagulation and transection cycles were required per ovarian pedicle. Intraoperative hemorrhage was minimal and only occurred if the mesovarium was transected in an area before adequate coagulation. Transection of the right ovarian artery in 1 horse without appropriate coagulation resulted in excess hemorrhage; the hemorrhage was quickly controlled by grasping the cut end of the artery with the electrosurgical forceps and then cauterizing the vessel. Repeat coagulation was required less than 3 times for all ovarian pedicles.

None of the fillies showed any signs of colic or inappetance after laparoscopic ovariectomy. All had mild swelling at the laparoscopic portal sites, but these regions were not painful on palpation. No incisional complications occurred in any of the fillies.

Four of the ovaries were found free-floating within the abdomen. The other 16 ovaries were enveloped in the free portion of the greater omentum, adjacent to the spleen, in the cranioventral abdomen. All the ovaries had brownish discolouration throughout the parenchyma of the cut sections and appeared to have multiple old follicles. The areas of transected mesovarium and associated uterine horn showed no adhesions or signs of inflammation on gross examination. There were no adhesions directly related to the surgical procedure except in the horse with visceral puncture.

Histologic examination of the attached ovaries consistently showed a band of fibrous tissue, interpreted to be omentum, surrounded the ovary. In all ovaries, the superficial parenchyma contained many mononuclear cells laden with golden pigment. Areas of the tunica albuginea, and some superficial stromal cells, appeared viable. The deep stromal cells were partially mineralized, and all

deep blood vessels and all examined follicular structures were necrotic and partially mineralized. The attached and free-floating ovaries had similar histologic changes in the deep vasculature and follicular structures.

## Discussion

Ventral abdominal laparoscopy was performed in these fillies because of their smaller size (standing procedures would be more difficult) and the relatively unhandled nature of the group before the study. There should be no reason the same procedure could not be undertaken with the horse standing, as previously described (9–12). The standing horse, if cooperative, could minimize the added challenge of proper orientation of the ovary before coagulation and transection: suspension of the ovary from its dorsal attachments would allow excellent access to the mesovarium for coagulation. Standing surgery would also decrease the risks and expense of general anesthesia.

The mesovarium, once the ovarian bursa is removed, is divided into a thick medial component and a thin lateral component (12). During our operations, both components were cauterized and transected. Because the medial segment contains the vascular supply of the ovary, other authors have performed coagulation and transection of the medial segment and simply transected the lateral component, without any complications (12). Operative time could be reduced if this technique were used.

Electrosurgical coagulation and transection of the ovarian pedicle was chosen for several reasons. First, electrocoagulation is a reliable means of mesovarium hemostasis in the horse (12). It was also an economical alternative: other techniques necessitate the purchase of costly consumable materials. Confidence in the degree of coagulation. Standing surgery before transection increased greatly with the number of surgical procedures. Other instruments that we considered for coagulation included the recently described bipolar vessel-sealing device (15) and the tripolar cutting forceps (Kendall; Tyco Healthcare Group LP, Mansfield, Massachusetts, USA). The feedback-controlled bipolar vessel-sealing device provides excellent hemostasis while precise energy is applied to seal the vessels; however, it was considered economically unfeasible at the time. The tripolar instrument was not available.

The use of only 2 instrument portals was feasible because of the ease with which the electrosurgical forceps could be applied to the mesovarium and exchanged with the laparoscopic scissors via 1 portal. Limiting the number of portals decreased the time required to suture the abdomen closed and theoretically decreased patient morbidity. The described techniques for multiple working portals may be more appropriate for the removal of unilateral diseased ovaries, such as those with granulosa cell tumours (8).

An unexpected result of the study was the noticeable lack of abdominal discomfort and inappetance among the fillies after surgery. The use of flunixin meglumine, 0.5 mg/kg, could be expected to give some degree of analgesia (21). Multiple studies have found that mares undergoing laparoscopic ovariectomy sporadically exhibit signs of colic in the face of a higher dose of flunixin (1.1 mg/kg) or phenylbutazone (4.4 mg/kg, given orally every 12 h) (8–10,12). In a large proportion of these previously described horses, a local anesthetic was injected into the ovarian pedicle before transection. In our experience, postlaparoscopic colic seems to be limited to ovariectomy patients, whereas horses undergoing cryptorchidectomy and exploratory laparoscopy rarely exhibit postoperative pain. The addition of butorphanol tartrate (0.05 mg/kg, given intravenously at the start of the procedure) should also aid in short-term analgesia (22), but long-term medication for pain control was not necessary. The nerves associated with the ovaries are derived from the sympathetic system through the renal and abdominal aortic plexuses (23). To our knowledge, no study has examined the development of the ovarian nervous supply in the maturing horse. Younger animals may have incomplete or less developed innervation of the ovarian pedicle. Although we studied only 10 juvenile horses, our findings suggest that patient morbidity might be decreased if ovariectomy were performed in younger animals. In addition, the juvenile vascular structures are smaller, and hemostasis may be more readily achieved.

The adhesion between the tip of the cecum and the omentum in 1 filly was not surprising. The location of the adhesion and the fact that intestinal contents were seen in the hub of the insufflation needle at the time of surgery suggest that the teat cannula was placed in the cecum. Before the filly's hindquarters were elevated for surgery, the entry point of the teat cannula was not observed with the laparoscope. Sufficient insufflation and preoperative fasting should decrease the risk of inadvertent puncture of a viscus. Careful insertion of the teat cannula through the peritoneum will also decrease the chance of visceral damage.

Similarly, the attachment of a large proportion of the ovaries to the omentum can be rationalized. Gravity would tend to displace the unattached ovaries to the cranioventral aspect of the abdominal cavity. The omentum is particularly prone to adhesion formation because it can move freely throughout the peritoneal cavity (24). The omentum and its ability to cover serosal defects have been extensively described (25,26). It has been experimentally demonstrated that the omentum will adhere to ischemic bowel within 48 h of contact (25). Contributing to adhesion formation is the fact that the entire surface of the ovary is not covered with a mesothelial layer: the dorsal or convex surfaces, where the vessels and nerves enter, are devoid of a peritoneal lining (27). Coagulation and transection exposes the cut edge of the mesovarium and the portion of the ovary without a peritoneal lining while causing local inflammation and ischemia. Adherence of the ischemic ovaries to the free portion of the greater omentum in the ventral abdomen is likely if the exposed connective tissue is unable to repair itself with mesothelial cells. Ischemic bowel adhering to omentum undergoes neovascularization 3 to 4 d after attachment (25). Since, as we observed histologically, the deep vascular supply and all follicular structures of the ovaries were nonviable, attachment of the omentum to the ovaries and neovascularization must have occurred at a rate incompatible with cell survival. In contrast, in feline species in which ischemic ovaries were surgically transected and implanted into the small intestinal mesentery in the abdomen, the ovarian tissue attached and revascularized, producing measurable hormone levels, before complete cell death (28). The difference in this study, other than species, was

that the ovaries were tethered to the mesentery with suture and sectioned before implantation. This eliminated the time required for the ovaries to come into contact with the omentum and, therefore, decreased the time required for revascularization of the ovarian tissue. Similarly, complete transection of the mesovarium and not the ovarian vessels alone was deemed necessary to promote avascular necrosis of the ovary. In situ coagulation and transection of the testicular cord of cryptorchid testicles without removal from the inguinal ring resulted in revascularization and subsequent production of testosterone (29).

Unlike the ovaries of other domestic mammals, equine ovaries have a peripheral zone of connective and vascular tissue around a central parenchymatous zone containing developing and atretic ovarian follicles, corpora lutea, and corpora albicantia (30). Transection of the ovarian pedicle removes the blood supply to the ovary. Revascularization by the omentum, if rapid enough, could provide the ovarian cells with a new blood source before avascular necrosis. The fact that the structures responsible for hormone production and follicle development are deep in the ovaries of the horse may account for the difference observed between the cat and the horse. It is plausible that the deeper structures of the transected ovaries underwent cellular death and necrosis before revascularization.

Hormonal assays were not performed in this group of horses because of their age (4 to 5 mo). Puberty in the horse is generally accepted to occur between 12 and 24 mo (31), and for that reason serum hormonal analysis in these horses could not be relied upon. The procedure needs to be repeated in mature, actively cycling mares and the behavioural and hormonal effects analyzed.

The envelopment of the majority of the transected ovaries in the free portion of the greater omentum brought about the question of potential complications. The possibility of the attached ovaries acting like a mesenteric lipoma and causing strangulation of a portion of bowel was considered. The fact that the ovaries were enveloped in the omentum and did not have a stalk decreased the likelihood of strangulation. The gravitational force associated with a suspended abdominal mass is generally regarded as a major factor in the development of a stalk. In our horses, the enveloped ovaries lay in the cranioventralmost aspect of the abdomen, such that stalk development was unlikely.

Our results were encouraging, and use of this surgical procedure to control undesirable estrus behaviour in the mare should be considered. In the young, acyclic horse, laparoscopic ovariectomy involving coagulation and transection of the ovarian pedicle without removal of the ovaries from the abdominal cavity appears to be an alternative to other ovariectomy techniques. There was no postoperative morbidity, no peritoneal adhesions were associated with the dropped ovaries, and avascular necrosis was achieved in all the deep blood vessels and all follicular structures of the coagulated and transected ovaries.

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