Technical Note: Improved technique for fitting pigs with steered ileocecal valve cannulas

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ABSTRACT: Collection of ileal digesta to evaluate AA digestibilities has become increasingly important in swine nutrition research. Steered ileocecal valve cannulation of pigs permits total collection of ileal digesta, while still allowing normal digesta flow during noncollection periods. This technique was modified and used with 64 crossbred barrows in five trials. Our procedural changes included preoperative i.v. administration of a broad-spectrum antibiotic and nonsteroidal antiinflammatory drug, sharp incision through the muscle layers of the laparotomy wound, use of a heparinized saline lavage solution, replacement of the guide ring with a stylette, and fixing the outer cannula barrel in place with a hose clamp. The current technique involves a right flank laparotomy, parallel and distal to the last rib, with the pig under general anesthesia. A stainless-steel ring (inner ring = 2.0 mm thick, 35.0 mm i.d.) is introduced into the ileal lumen through an enterotomy proximal to the origin of the ileocecal fold. A nylon string attached to this ring is threaded through the ileum and ileocecal valve into the cecum using a silastic stylette, which encases the string. A second stainless-steel ring (outer ring = 2.0 mm thick, 34 mm o.d.) is fixed in place around the ileum, distal to the inner ring and just proximal to the ileocecal valve. A polyurethane cannula barrel (barrel = 100 mm long, 26 mm i.d., 32 mm o.d.; flange = 70 mm o.d.) is introduced into the cecal lumen via an enterotomy through the lateral cecal band and secured in place with two purse-string sutures. The cannula is exteriorized through an incision caudal and proximal to the intial laparotomy site, where it is plugged using a cylindrical stopper (26 mm o.d., 55 mm long) and held in place by a second cannula barrel (barrel = 43 mm length, 33 mm i.d., 41 mm o.d.; flange = 80 mm o.d.). Procedural changes decreased postsurgical complications, as evidenced by decreased seepage around the cannula and fewer and less severe adhesions noted at necropsy. Based on five trials, this technique is a reliable means of collecting ileal digesta for nutrient analyses.

Key Words: Amino Acid, Cannula, Cecum, Ileum, Pigs, Surgery


Introduction

With growing emphasis on the ileal digestibility of nutrients, it is essential that a reliable method for collecting ileal digesta is developed. Currently, ileal digesta is collected using one of three methods: slaughter technique, ileorectal anastomosis, or with a cannulated pig. The slaughter technique is relatively simple; however, it has several disadvantages, including the inability to obtain multiple samples per animal, difficulties in obtaining representative samples, and animal-to-animal variation. Ileorectal anastomosis involves removal or complete bypass of the cecum and colon; and digesta samples are collected from the anus. This procedure provides an easy method for collecting digesta, but unfortunately, the normal digestive state is not maintained (Albin et al., 1999). In addition, the removal of such a large portion of the gastrointestinal tract may alter gut physiology (Kohler et al., 1992). Several cannulation techniques for collection of ileal digesta exist, including: 1) simple T-cannulation; 2) postvalvular T-cannulation; and 3) reentrant cannulation. Nonetheless, these techniques do not allow for quantitative sampling; therefore, one must rely on a marker for digestibility measurements. In addition, these cannulas may become plugged or alter the normal passage of digesta, particularly in high-fiber diets. Mroz et al. (1996) imple-
mented a new cannulation technique called steered ileocecal valve (SICV) cannulation (Figure 1). This procedure allows for quantitative collection of ileal digesta, but requires a more complex surgery, allowing greater opportunity for postsurgical complications. The objectives of these experiments were to 1) improve the surgical technique for inserting the SICV cannula, thereby minimizing postsurgical complications; and 2) improve the cannula design to minimize leakage around the cannula.

**Materials and Methods**

**Description of Cannula**

The SICV cannula comprises five parts (Figure 2): an inner cannula barrel (polyurethane; barrel = 100 mm long, 26 mm i.d., 32 mm o.d.; flange = 70 mm o.d.), an outer cannula barrel (polyurethane; barrel = 43 mm long, 33 mm i.d., 41 mm o.d.; flange = 80 mm o.d.), an internal ring (stainless steel; 2.0 mm thick and 35.0 mm i.d.) attached to a nylon cord, an external ring (stainless steel; 2.0 mm thick and 34 mm o.d.), and a cylindrical stopper (polyurethane; 26 mm o.d. and 55 mm long).

**Surgical Preparations**

All procedures were approved by the University Animal Care and Use Committee. Pigs were approximately 35 kg BW at the time of surgery. Feed and water were withheld for 36 and 12 h before surgery, respectively.

General anesthesia was induced with a combination of medetomidine (80 μg/kg of BW), ketamine (10 mg/kg of BW), and butorphanol (200 μg/kg BW) given intramuscularly and maintained with isoflurane and oxygen via an endotracheal tube through a semiclosed circle anesthetic system. Immediately after induction of anesthesia, ceftiofur (Naxcel, 2.2 mg/kg of BW, Pfizer, New York, NY) and flunixin meglumine (Banamine, 1.1 mg/kg BW, Schering-Plough Animal Health Corp., Kenilworth, NJ) were administered intravenously. During surgery, a balanced electrolyte solution (2 to 5 mL/kg of BW/h) was administered through a catheter placed in an ear vein. Pigs were positioned in left lateral recumbency and prepared for aseptic abdominal surgery. Members of the surgical team scrubbed, gowned, and gloved routinely. The surgical site was draped with a sterile impervious sheet. The surgeon’s gloves were thoroughly rinsed with sterile saline.

**Surgical Procedures**

A 10-cm skin incision was made distal (approximately 2 to 3 cm) and parallel to the last rib. The incision was continued sharply (i.e., with a scalpel as opposed to blunt dissection) through the s.c. tissue, cutaneous muscle, external and internal abdominal oblique muscles, transverse abdominal muscle, and peritoneum. Bleeding was controlled by application of hemostats and/or ligation.

The cecum was identified and retracted dorsally to expose the terminal ileum and ileocecal junction. The cecum and ileum were isolated from the abdominal cav-
Figure 2. Parts of the steered ileocecal valve cannulation system: A) an outer cannula barrel (barrel = 43 mm long, 33 mm i.d., 41 mm o.d.; flange = 80 mm o.d.); B) external ring (2.0 mm thick and 34 mm o.d.); C) inner cannula barrel (barrel = 100 mm long, 26 mm i.d., 32 mm o.d.; flange = 70 mm o.d.); D) cylindrical stopper (26 mm o.d. and 55 mm long); and E) internal ring (2.0 mm thick and 35.0 mm inner diameter) attached to a nylon cord.

ity with heparinized (10 U/mL) saline-soaked sponges. A 3.5-cm enterotomy was made on the long axis of the ileum along the antimesenteric border, just proximal to the origin of the ileocecal fold. The inner ring and attached nylon cord were introduced into the lumen of the ileum using a silastic stylette. The stylette was manipulated through the ileocecal valve and into the cecum. The enterotomy was closed with 3-0 polydioxanone sutures using a continuous Cushing pattern. Following lavage of the enterotomy site with heparinized saline and regloving by the surgeons, the outer ring of the steering system was placed around the ileum close to the ileocecal junction. This ring was cut so that it could be passed around the ileum and through the ileocecal mesentry. The ring was reconnected using a specially designed connecting crimp. The ileum was replaced into the abdominal cavity and the cecum was isolated with heparinized saline-soaked sponges.

A purse-string suture using 2–0 polydioxanone was preplaced around the lateral band of the cecum (approximately 8 cm long) as close to the level of the ileocecal junction as possible. An enterotomy was made through the lateral band of the cecum between the purse-string sutures. The stylette and nylon cord attached to the inner ring were retrieved from the cecum with forceps and threaded through the inner cannula barrel. The flange of the inner cannula barrel was then positioned in the cecum, and the purse string suture tightened and tied. A second purse string suture was placed around the cannula for added security. The silastic stylette was carefully removed from the nylon cord, and the nylon cord was incorporated into a gauze plug, which was placed in the inner barrel to prevent the outflow of digesta.

Following lavage of the enterotomy site and regloving by the surgeons, the inner cannula barrel was exteriorized through a wound created caudal and proximal to the laparotomy site. This wound was created by removing an approximately 3-cm-diameter section of skin and then bluntly “gridding” through the abdominal musculature. The wound was positioned so that the flange of the cannula barrel did not interfere with the musculature of the hindlimb or contact the kidney. The cannula was exteriorized and pulled snugly against the body wall. The inner cannula barrel was plugged (after removing the gauze plug) with a cylindrical stopper and secured in place by the outer cannula barrel with a stainless-steel hose clamp and plastic wire tie. Abdominal closure was performed in four layers. The transverse abdominal muscle was closed with 2–0 polydioxanone...
none in a simple continuous pattern. The peritoneum was closed with the transverse abdominal muscles if it were easily accessible, but in many cases it was not. In our experience, we observed no differences in postsurgical recovery with or without closure of the peritoneum. The internal and external abdominal oblique muscles were closed together using 0 polydioxanone in a simple continuous pattern. The cutaneous muscle and s.c. fascia were closed together using 3-0 polydioxanone in a simple continuous pattern. The skin was closed with 3–0 polydioxanone in a continuous subcuticular pattern. Throughout the surgical procedure, the heparinized saline solution was used to wash the surgeons’ gloves, to keep tissues moist, and to lavage the enterotomy sites.

**Animals, Housing, and Postsurgical Care**

Administration of cefiofur (2.2 mg/kg of BW i.m. every 12 h) and flunixin meglumine (1.1 mg/kg of BW i.m. every 24 h) was continued for 72 h after surgery. Pigs were individually housed in specially designed metabolism pens (1.2 × 1.2 m). On the day after surgery, pigs were fed 50 g of a standard corn-soybean meal diet in two daily feedings (0600 and 1800). Feed intake was increased 100 g/d up to 9% of metabolic BW (BW0.75), and water was supplied ad libitum. The area around the cannula was cleaned as needed with warm water, and outer cannula barrels were adjusted weekly to accommodate growth.

**Results and Discussion**

During the development of this technique, 64 pigs, over five different experiments, were cannulated. One pig was killed 4 wk after surgery from the first surgery set due to lack of appetite and sudden weight loss. At necropsy, a jejunal intussusception, blocking the flow of digesta, was observed. Of the remaining 63 pigs, all were used in experiments (8 to 6 wk long) without complications. Surgical procedures used in Exp. 1 were identical to those described by Mroz et al. (1996). Pigs (n = 12) used in this experiment were necropsied 18 wk after surgery. Mild to severe adhesions involving the distal small intestine were observed in 50% of the pigs. It was speculated that adhesion formation was potentiated by lack of preoperative antibiotic and nonsteroidal antiinflammatory therapy, excessive manipulation of the bowel during placement of the internal ring, accumulation of fibrinous exudates, dried blood on the surface of the bowel and within the abdominal cavity, and contamination (glove powder, bacteria, and/or ingesta from the enterotomy sites).

In later surgeries, actions were taken in an attempt to prevent/minimize adhesions, including 1) implementation of preoperative nonsteroidal antiinflammatory and broad-spectrum antibiotic therapy; 2) alteration of the laparotomy approach and intraabdominal techniques; and 3) use of a stylette to replace the guide ring described by Mroz et al. (1996) for passage of the string from the internal ring through the ileal lumen and into the cecum.

Perioperative treatment with broad-spectrum antibiotics and nonsteroidal antiinflammatory drugs has been shown to decrease postoperative peritoneal adhesions, presumably by decreasing peritoneal inflammation and intraperitoneal bacterial load (Southwood and Baxter, 1997; Sullins et al., 2004). It is important that therapy begin preoperatively (before peritoneal inflammation/contamination occurs) for maximum benefit. In Exp. 1, antibiotic and nonsteroidal antiinflammatory therapies were not instituted until after completion of surgery. In subsequent experiments, treatment was begun (by i.v. administration) shortly after induction of anesthesia.

In the work of Mroz et al. (1996), the laparotomy approach involved incision through the skin and s.c. tissue, and then blunt dissection or “gridding” through each muscle layer (muscle layers divided along the direction of the muscle fibers). This technique was used in Exp. 1. A grid approach results in a smaller functional surgical field than a similarly sized approach involving vertical incision through all tissue layers. In Exp. 2 to 5, a vertical incision approach was used. This technique resulted in better exposure and decreased the amount of bowel handling/manipulation that would predispose the intestine to adhesion formation. In particular, less traction was needed around the ileocecal junction while the string attached to the inner ring was threaded through the ileal lumen and into the cecum.

A variety of stylette designs were tried throughout the experiments, but the most effective was a silastic tube that encased the string attached to the inner ring with a stainless steel head attached at the end. Use of the stylette decreased manipulation of the bowel during insertion of the string and ring system into the ileal lumen and cecum. The stainless-steel head allowed easier direction of the stylette through the ileocecal valve and into the cecum.

Other techniques used in Exp. 2 to 5 in an attempt to prevent adhesions included keeping the bowel moist at all times to prevent drying and to minimize friction during manipulation, thorough lavage of the enterotomy sites after closure to remove fibrinous exudates, blood, debris and bacteria, and mandatory regloving and washing of the gloves after closure of the enterotomies. Heparin was added to the lavage solution in attempt to decrease the conversion of fibrinogen to fibrin, and thereby decrease adhesion formation. Mandatory regloving and glove washing was implemented to decrease bacterial contamination and introduction of foreign material (glove powder) into the abdomen.

Before modifications of the procedures described by Mroz et al. (1996), severe adhesions, defined as visible adhesions capable of altering normal digesta flow, were observed in 20% of the pigs that were necropsied (n = 10). Moderate adhesions, defined as mild to moderate adhesion formation with no visible impairment of digesta flow, were observed in an additional 30% of the
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pigs. After modifying the procedure as described above, severe adhesions were not observed in any pigs (n = 52), and moderate adhesions were observed in less than 16% of all pigs in each experiment (average = 13%).

Leakage around the cannula barrel was decreased by the addition of a tapered barrel to the cannula, and by the use of a stainless-steel hose clamp to secure the outer barrel in place. Adding a taper to the inner cannula barrel allowed for exteriorization of the cannula through a smaller wound site in the body wall, which decreased seepage around the cannula. The outer cannula barrel was held tightly in place by the use of a stainless steel hose clamp. This minimized movement of the cannula through the body wall, decreasing expansion of the opening, thereby decreasing seepage.

In summary, SICV cannulas allow for a total collection of ileal digesta, thereby eliminating the variation of representative sampling techniques. This procedure, developed by Mroz et al. (1996), allows for digesta to flow normally when it is not being collected, and it does not require digesta to pass through the cannula barrel. This eliminates the threat of digesta blockage by the cannula, particularly when feeding high-fiber diets. Improvements made to the original procedure described by Mroz et al. (1996) minimized complications after surgery and included 1) modification of the abdominal approach to allow a larger working field and therefore greater access to the bowel; 2) regloving and using heparinized saline lavage to decrease contamination, to prevent fibrin accumulation, and to keep the bowel moist; 3) use of a stylette for insertion and placement of the internal ring to minimize handling of the bowel; 4) redesign of the inner cannula barrel to include a taper that eased passage through the body wall; and 5) fixing the outer cannula barrel in place with a stainless-steel hose clamp to minimize digesta leakage around the cannula. These alterations to the original technique have decreased postsurgical complications, yielding a viable research tool for collection of ileal digesta.

Literature Cited


