In vitro comparison of leakage pressure and leakage location for various staple line offset configurations in functional end-to-end stapled small intestinal anastomoses of canine tissues

Lane A. Hansen DVM, MS
Daniel D. Smeak DVM

Received April 12, 2014.
Accepted December 22, 2014.

From the Military Working Dog Center Europe, Pulaski Barracks, Kaiserslautern, Germany (Hansen); and the Department of Clinical Sciences, College of Veterinary Medicine and Biomedical Sciences, Colorado State University, Fort Collins, CO 80523 (Smeak).

Address correspondence to Dr. Smeak (dan.smeak@colostate.edu).

OBJECTIVE
To compare intraluminal pressure at initial leakage (leakage pressure), leakage location, and maximum intraluminal pressure (MIP) for various staple line offset configurations of functional end-to-end stapled anastomosis (FEESA).

SAMPLE
Grossly normal jejunal segments from 4 canine cadavers.

PROCEDURES
52 jejunal segments (4 control and 24 anastomosis constructs [2 segments/standard FEESA construct]) were prepared for testing. Segments were assigned to three 8-segment gastrointestinal anastomosis staple line offset groups: complete offset (CSO group), partial gastrointestinal anastomosis staple line offset (PSO group), and no gastrointestinal anastomosis offset (NSO group). Results for leakage pressure, leakage location, and MIP were compared.

RESULTS
Mean ± SD leakage pressure differed significantly among all groups and was highest for the PSO group (34.4 ± 3.7 mm Hg), followed by the CSO group (25.9 ± 4.1 mm Hg) and the NSO group (18.8 ± 1.5 mm Hg). Leakage location did not differ significantly among groups but was most commonly associated with the thoracoabdominal staple line. The MIP did not differ significantly among groups (PSO, 83.1 ± 9.4 mm Hg; CSO, 81.7 ± 6.7 mm Hg; and NSO, 58.5 ± 7.7 mm Hg).

CONCLUSIONS AND CLINICAL RELEVANCE
In this study, partial staple line offset leaked at a significantly higher pressure, which represented the greatest leakage protection of tested constructs. The thoracoabdominal staple line was more susceptible to leakage than was the gastrointestinal anastomosis staple line. Results suggested that surgeons should avoid FEESA with no staple line offset, strive for partial offset of the gastrointestinal anastomosis staples, and provide precise placement of the thoracoabdominal staple line. (Am J Vet Res 2015;76:644–648)
blood flow, and anastomosis failure. Staple position, staple intersection, and staple method may be mechanical factors that influence performance of FEESA constructs.

The purpose of the study reported here was to evaluate leakage pressure, leakage location, and MIP for FEESA constructs and to compare results with those reported for other studies conducted to evaluate sutured anastomoses tested by use of the same methods. Our primary hypothesis was that stapled anastomoses would have leakage and MIP values similar to those reported for sutured anastomoses regardless of the configuration of the gastrointestinal anastomosis staple line used for closure of the terminal end. Our secondary hypothesis was that there would be no difference in leakage location between the thoracoabdominal and gastrointestinal anastomosis staple lines.

Materials and Methods

Sample

Segments of jejunum were harvested from 4 similar-sized purpose-bred large-breed research dogs euthanized for reasons unrelated to the study. The dogs did not have clinical signs of disease, and their small intestines appeared grossly normal. Dogs were euthanized by IV infusion of pentobarbital or phenytoin sodium. Jejunum (from aborad to the caudal duodenal flexure to the ileum) were harvested immediately after the dogs were euthanized. Intestines were cut with Mayo scissors into 40- to 60-cm segments, placed in sterile saline (0.9% NaCl) solution, and stored at 4°C until used in an anastomosis construct; all segments were used within 24 hours after harvest. Immediately before use in testing, segments were removed from the saline solution and cut into 6- to 8-cm sections for use in the anastomosis constructs. Luminal contents were manually milked out of these sections (similar to preparation of the bowel for resection and anastomosis).1

Anastomosis construction

Fifty-two intestinal segments were used in the study. Segments were assigned to experimental groups by use of a randomization procedure (segments were randomly chosen and assigned in a rotating order to the 4 groups until all groups were complete). Four segments were selected for validation of the pressure testing procedure and assessment of intestinal integrity (control group). The remaining segments were allocated into 3 groups (16 segments/group, consisting of 2 segments/construct and 8 constructs/group): NSO group, PSO group, and CSO group (Figure 1). All groups (except the control group) were prepared for anastomosis constructs by fully seating an intestinal segment on each limb of the gastrointestinal anastomosis stapling device, which was loaded with a 3.8-mm staple cartridge as described elsewhere.

The limbs of the stapler were engaged and locked, while ensuring that the jejunal segments remained fully seated (with the mesenteric border of each segment approximately parallel to each other and on the center of the outer faces of the stapling device). The stapling device was triggered, which placed 2 double-staggered staple rows through both segments. The cutting assembly was advanced along the stapling device to create a stoma between the segments.

The gastrointestinal anastomosis staple lines were identified and oriented for each group, and staples from a thoracoabdominal stapling device loaded with a 3.5-mm staple cartridge were applied across the intestinal edges approximately 1 cm from the transverse opening. For the NSO group, a stay suture was inserted 2 to 3 mm from each terminal end equidistant between gastrointestinal anastomosis staple lines. Tension was placed on the stay sutures, which ensured that the gastrointestinal anastomosis staple lines were directly overlapping; the thoracoabdominal stapler then was closed on the tissue. For the PSO group, a stay suture was inserted 2 to 3 mm from each terminal end at approximately 60° counterclockwise from the gastrointestinal anastomosis staple line. Tension was placed on the stay sutures, which created an offset of the staple lines of approximately 2 cm; the thoracoabdominal stapler then was closed on the tissue. For the CSO group, a stay suture was inserted 2 to 3 mm from each terminal end and through each gastrointestinal

Figure 1—Photographs of staple position for the NSO group (A), PSO group (B), and CSO group (C and D) in jejunal sections obtained from canine cadavers. For the CSO group, gastrointestinal anastomosis staple lines are at the left and right sides near the stay sutures (panel C) and are indicated (black circles) on the completed construct (panel D).
anastomosis staple line. Tension was placed on the stay sutures so that they were oriented 180° to each other and the intestinal tissue was not stretched; the thoracoabdominal stapler then was closed on the tissue. After appropriate positioning was completed for all groups, the thoracoabdominal stapler was triggered. Excess tissue (which included the tissue containing the stay sutures) was trimmed with curved Mayo scissors, and the stapler was released. A single interrupted suture (4-0 glycomer 631 swaged onto a CV-23 half-circle, 17-mm tapered needle) was placed through the submucosa of both segments at the notch formed on the anti-mesenteric border where the crotch suture is placed.

Methylene blue–saline solution was delivered at a rate of 999 mL/h with an infusion pump. Completed constructs were placed in saline solution to control tension and prevent separation of the 2 segments. Completed constructs were placed in saline solution until all anastomoses were performed. At the end of the construction session, all constructs were stored at 4°C until testing was performed.

**Pressure testing**

Testing was performed the day the anastomoses were constructed; thus, all constructs were stored for < 24 hours. Testing was performed as described elsewhere for sutured anastomosis constructs. Jejunal constructs and the control group (which was tested to validate the testing technique and health of the intestinal tissues) were tested in random order. A 4-cm-long, 9F catheter was inserted to a depth of 3.5 cm into 1 end of a section, which was then sealed with a Doyen clamp placed from the mesenteric border as close as possible to the catheter (approx 3 cm from the catheter tip; Figure 2). A right-angle Rochester-Carmalt forceps was tightly placed parallel to the catheter and perpendicular to the Doyen clamp to prevent leakage. A microtip pressure transducer was introduced to a depth of 3 to 4 cm into the lumen at the opposite end of the section. The transducer, which was calibrated before each experiment, was connected to a data acquisition system. The transducer was secured with a Doyen clamp placed perpendicularly across the intestine and transducer. The instrumented section of intestine was then submerged in a plastic tub filled with water. An infusion catheter system was primed with methylene blue in saline solution (dilution, 1:500). The methylene blue–saline solution was delivered at a rate of 999 mL/h with an infusion pump.

The system was purged of air, recording of pressure measurements was begun, and infusion of the methylene blue–saline solution was started. Leakage pressure and location of leakage were recorded. Leakage pressure was defined as the pressure at which the methylene blue–saline solution was first observed leaking from a construct. Once leakage pressure was recorded, testing to determine MIP continued until there was catastrophic failure of the anastomosis or rupture of the intestinal tissue, intraluminal pressure reached a plateau, or maximum pressure for the sensor system was achieved.

**Statistical analysis**

Statistical analysis was performed with commercially available software. Results were reported as mean ± SD. A 1-way ANOVA was performed to evaluate results, and the Student’s t test was used to compare results among groups. Values of P ≤ 0.05 were considered significant.

**Results**

Testing of the control sections revealed a mean ± SD leakage pressure of 250.5 ± 46.6 mm Hg. Mean leakage pressure was 34.4 ± 3.7 mm Hg for the PSO group, 25.9 ± 4.1 mm Hg for the CSO group, and 18.8 ± 1.5 mm Hg for the NSO group. Leakage pressure differed significantly among groups; leakage pressure was highest (P = 0.04) for the PSO group and lowest (P < 0.001) for the NSO group.

Leakage location was limited to the thoracoabdominal staple line for 17 of 24 (71%) constructs, was identified at or near the intersection of the thoracoabdominal and gastrointestinal anastomosis staple lines for 5 of 24 (21%) constructs, and was located at the suture-buttressed region of the gastrointestinal anastomosis staple line with concurrent leakage at the thoracoabdominal staple line for 2 of 24 (8%) constructs. In constructs that had gastrointestinal anastomosis staple line failure, that failure consistently occurred at the end of the gastrointestinal anastomosis staple line opposite from the thoracoabdominal staple line. There was no significant (P = 0.33) difference in leakage location among groups; however, whereas thoracoabdominal staple lines leaked without concurrent leakage of the gastrointestinal anastomo-
sis staple line, there were no gastrointestinal anastomosis staple lines that leaked without concurrent leakage of the thoracoabdominal staple line. The thoracoabdominal staple line leaked most commonly at or in the proximity of (within 2 to 3 mm) the intersection of the thoracoabdominal staple line and the gastrointestinal anastomosis staple line.

The MIP did not differ significantly ($P = 0.07$; 1-way ANOVA) among groups. Mean ± SD MIP was $83.1 \pm 9.4$ mm Hg for the PSO group, $81.7 \pm 6.7$ mm Hg for the CSO group, and $58.5 \pm 7.7$ mm Hg for the NSO group.

**Discussion**

Use of an efficient and effective leak-proof technique for treating leaking, damaged, or devitalized bowel is paramount in the removal and treatment of such tissue. The purpose of the study reported here was to evaluate positioning of gastrointestinal anastomosis and thoracoabdominal staple lines on the performance of FEESA repairs to identify factors related to technique that affect staple line performance. The PSO group had the highest leakage pressure, compared with leakage pressures for the CSO and NSO groups. These results led to a partial rejection of the primary hypothesis that staple line configuration did not influence leakage pressure of stapled anastomoses. However, we found that stapled anastomoses had similar leakage pressure, compared with results for sutured anastomoses reported previously. Thus, we partially accepted the primary hypothesis that staple line configuration did not influence leakage pressure, compared with results for sutured anastomoses. Mean ± SD leakage pressure for the PSO group ($34.4 \pm 3.7$ mm Hg) was similar to that for a group sutured with 3-0 USP glycomer 631 ($34.0 \pm 6.9$ mm Hg) reported in another study. Both of these results were greater than mean leakage pressure for a group sutured with 4-0 USP glycomer 631 ($28.0 \pm 6.7$ mm Hg); results for the group sutured with 4-0 USP glycomer 631 in that study were similar to the mean leakage pressure for the CSO group ($25.9 \pm 4.1$ mm Hg) in the present study.

Mean ± SD leakage pressure for the NSO group ($18.8 \pm 1.5$ mm Hg) revealed a possible danger for leakage of these anastomoses during peristalsis or prior to the use of other techniques. Intraluminal pressure of the small intestines is 15 to 25 mm Hg during peristalsis in healthy dogs. The anastomosis groups in the study reported here leaked at pressures similar to those for sutured anastomoses reported elsewhere, although the NSO group leaked at pressures within the ranged expected during peristalsis. Analysis of results of the present study suggested that the PSO technique provided an increased margin of leakage protection, compared with that for the NSO or CSO groups and for sutured anastomoses. Leakage at a lower pressure may have been related to a lack of depth of penetration for staples of the thoracoabdominal staple (because of greater tissue mass) or interference with gastrointestinal anastomosis staples when the staple lines directly overlapped (NSO) or intersected at a right angle (CSO) when completely offset, as was noted in another study.

The MIP achieved in the present study was approximately half the pressure reported for suture anastomoses (FEESA group means between 58.5 and 83.1 mm Hg vs suture anastomosis group means between 146.0 and 185.2 mm Hg). This may have been associated with a number of factors, such as the rate of infusion or volume of the FEESA construct, a greater surface area involved in the construct, or technique error, which could lead to a loss of pressure. The MIPs recorded were similar among the PSO, CSO, and NSO groups of the present study; however, they were substantially lower than the pressure identified for the suture anastomoses reported in another study, which caused us to partially reject the primary hypothesis that they would be similar. We do not believe that this result will have great clinical relevance because these supraphysiologic pressures are not likely to be reached in vivo and initial leakage would likely result in clinical signs associated with anastomosis failure.

Analysis of the results of the present study also revealed that the thoracoabdominal and gastrointestinal anastomosis staple lines behaved differently during testing. The thoracoabdominal staple line was more prone to leakage at or near the intersection of the 2 staple lines. This may have been the reason that the NSO group had a lower leakage pressure. Leakage of the gastrointestinal anastomosis staple line consistently occurred at the V (ie, crotch) area, even though the area was buttressed with a suture to counter tension that may have been present. We did not test constructs without this buttress suture because suture placement is part of the standard FEESA technique. Leakage could have been associated with failure of the buttress suture to control tension on the gastrointestinal anastomosis staple line or with size of the needle used to insert the suture (ie, if the hole created by the needle was not sufficiently filled by the suture). Future studies could be conducted to evaluate the value of a buttress suture and determine whether this area could be made more secure. Analysis of these results led us to reject the hypothesis that the 2 staple lines leaked at the same location because constructs with offset of the gastrointestinal anastomosis staple line behaved differently.

The amount of time needed for completion of each construct was not recorded. Only instruments and supplies available in a surgical setting were used in construct preparation and were clinically applicable. Although we did not test for efficiencies gained by repeated construct preparation, influence as a result of refinement of technique was accounted for through the random assignment of the order in which constructs were completed.

Limitations of the present study included the use of cadaveric tissue that may have behaved differently or held staples differently than intestinal tissues in live
dogs. The harvested intestinal segments were com-
mingled, and tissues from > 1 dog may have been used
to create the 2-part constructs. The nonphysiologic
testing method (tissues held at 4°C before and for up
to 24 hours after preparation before failure testing)
may not mimic physiologic conditions. Handling and
testing of the tissue were performed as described for
other experiments to enable us to compare results for
the present study with results for those experi-
ments. Blood supply or healing was not assessed; both
of those factors could be affected by use of an offset
placement technique, although no effect of stapling
on those factors has been reported. Histologic ex-
amination to investigate the interaction of staples or
the depth or placement of staples was not performed.

In the present study, the PSO modification was an
effective method for increasing leakage pressure of
in vitro FEESA constructs in tissues obtained from ca-
nine cadavers. The FEESA technique is widely used in
clinical practice; and the partial offset modification
reported here should be expected to perform similarly
in clinical cases.

Acknowledgments

The views expressed in this article are those of the authors and do not
reflect the official policy or position of the Department of the Army,
the Department of Defense, the US Government, or Coviden Animal
Health.

Footnotes

a. Beuthanasia-D Special, Intervet Inc, Merck Animal Health, Sum-
mit, NJ.
b. GIA 50 multiaxial stapling device, Tyco, United States Surgical,
Norwalk, Conn.
c. AutoSuture GIA 50 Premium, 3.8 mm, Tyco, United States Surgi-
cal, Norwalk, Conn.
d. TA 55 multiaxial stapling device, Tyco, United States Surgical,
Norwalk, Conn.
e. AutoSuture TA Premium, 3.5 mm, Tyco, United States Surgical,
Norwalk, Conn.
f. Biosyn, 40 USP, provided by Coviden Animal Health, Mans-
field, Mass.
g. FR 4, Boston Scientific Corp, Marlborough, Mass.
h. Micro-Tip catheter transducer, Millar Instruments Inc, Hous-
ton, Tex.
i. SonoLab, Sonometrics Corp, London, ON, Canada.
l. JMP, version 11, SAS Institute Inc, Cary, NC.

References

1. Tobias KM, Johnston SA. Small intestine. In: Veterinary surgery:
staples with sutures for anastomosis of the small intestine in
3. White RN. Modified functional end-to-end stapled intestinal
anastomosis technique and clinical results in 15 dogs. J Small
4. Tobias KM. Surgical stapling devices in veterinary medicine: a
5. Catena E, La Donna M, Gagliardi S, et al. Stapled versus hand-
sewn anastomoses in emergency intestinal surgery: results of a
anastomoses in emergency small intestine surgery: a retrospective
review of outcomes in a unique patient population. J Trauma
to-end stapled intestinal anastomosis and resection performed
by nonexpert surgeons for the treatment of small intestinal
intestinal tract anastomosis: incidence of postoperative comp-
lications, compared with the sutured anastomosis. Ann Surg
9. Leung TT, MacLean AR, Buie WD, et al. Comparison of stapled
versus handsewn loop ileostomy closure: a meta-analysis. J
10. Ravitch MM, Steichen FM. Techniques of stapling in the
11. Ullman SL, Pavletic MM, Clark GN. Open intestinal anastomosis
with surgical stapling equipment in 24 dogs and cats. Vet Surg
In: Fossom TW, ed. Small animal surgery. St Louis: Mosby,
2013;539–530.
13. Brolin RE, Ravitch MM. Studies in intestinal healing VI. Effect of
pharmacologically induced peristalsis on fresh intestinal anas-
14. Canalis F, Ravitch MM. Study of healing of inverting and everting
15. Nandakumar G, Richards BG, Trencheva K, et al. Surgical adhe-
sive increases burst pressure and seals leaks in stapled gastro-
16. Ravitch MM. Intersecting staple lines in intestinal anastomoses.
17. Ritchey ML, Lally KP, Ostericher R. Comparison of different
styles of stapled bowel anastomoses in a canine model.
18. Ziling T, Walther BS. Are intersecting staple lines a hazard in
19. Hansen LA, Monnet EL. Evaluation of a novel suture material
for closure of intestinal anastomoses in canine cadavers. Am J Vet
20. Hansen LA, Monnet EL. Evaluation of serosal patch supplemen-
tation of surgical anastomoses in intestinal segments from ca-
21. Tasaka K, Farrar JT. Intraluminal pressure of the small intesti-
te of the unanesthetized dog. Pflugers Arch 1976;364:35–44.
22. Merry CM, Shafi BM, Binyamin G, et al. Profiling surgical staplers:
effect of staple height, buttress, and overlap on staple line fail-