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Evaluation and validation of a paralumbar fossa trans-abdominal rumen fluid sampling technique

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Abstract

A simple method to obtain rumen fluid via the left paralumbar fossa was evaluated on 58 adult cattle. Using a 16-gauge 1.5 inch (3.8 cm) needle, rumen fluid was successfully collected and evaluated from 45 of 58 head (78%). The primary reason for failure was body wall thickness exceeding the 1.5 inch (3.8 cm) needle ($P = 0.0002$). Average rumen pH of the 45 cattle samples was 7.9; the test cattle were primarily fed guinea grass with occasional brewer's grain supplement. An average 3 protozoa per field under 40 x magnification were seen. No external hematomas or other swellings were seen during the 3-week follow-up observations, and no complications were noted. Eleven of the 58 cattle (19%) had increased body wall thickness, based on ultrasonographic follow-up. The average length of time required to obtain a rumen sample, measured by introduction of the needle to the withdrawal of the needle, was 3 seconds (range < 1 to 8 sec). We conclude that the left paralumbar fossa trans-abdominal rumen fluid sampling technique is a safe and efficient method to obtain rumen fluid in cattle with a moderately thin (≤ 30 mm) body wall.

Key words: cattle, rumen fluid, rumen evaluation, rumen sampling technique

Résumé

On a évalué une méthode simple pour recueillir le liquide ruménal à partir de la fosse paralombaire gauche chez 58 bovins adultes. Des aiguilles de calibre 16 de 1.5 pouces (3.8 cm) ont permis de recueillir efficacement le liquide ruménal chez 45 des 58 bovins (78%). L'insuccès venait surtout lorsque l'épaisseur de la paroi corporelle excédait la longueur de 1.5 pouces de l'aiguille ($P = 0.0002$). Le pH moyen du rumen chez les 45 bovins échantillonnés était de 7.9. Les bovins testés étaient nourris avec de l'herbe de Guinée et de la drêche occasionnellement. Le nombre moyen de protozoaires vus au grossissement de 40x était de 3 par champ. Il n'y a pas eu d'hématomes externes évidents ni d'autres enflures durant les trois semaines de suivi et aucunes autres complications n'ont été notées. Le suivi échographique a décelé une augmentation de l'épaisseur de la paroi corporelle chez 11 des 58 bovins (19%). Le temps moyen nécessaire pour recueillir un échantillon de liquide ruménal, mesuré depuis l'insertion de l'aiguille jusqu'à son retrait, était de 3 secondes (plage : 1 à 8 secondes). Nous concluons que la technique d'échantillonnage du liquide ruménal par la fosse paralombaire gauche à travers l'abdomen est un moyen sécuritaire et efficace de recueillir le liquide ruménal chez des bovins avec des parois corporelles modérément minces (≤ 30 mm).

Introduction

Evaluation of rumen fluid for pH and rumen microbes, in particular rumen protozoa, is helpful for both diagnostic purposes and treatment decisions. Determining the pH of rumen fluid can be useful to confirm acute ruminal acidosis as well as subacute ruminal acidosis. Evaluation of ruminal protozoa helps determine the necessity of ruminal transfaunation (the process of transferring rumen fluid from 1 ruminant to another). Protozoa will be dying or dead if a ruminant animal has been ill and off-feed for a rather short time, usually > 4 days.[1] Passage of an oro-ruminal tube is a means of obtaining a rumen fluid sample; however, this method may yield rumen fluid mixed with saliva, which may falsely elevate rumen pH. Tubing an animal also requires more time and effort than rumenocentesis. Nordlund and Garrett devised and tested a rumenocentesis method to diagnose subacute ruminal acidosis that utilized a 16-gauge, 5 inch (12.7 cm) needle, inserting the needle distal to the left paralumbar fossa.[4] A simpler method to collect rumen fluid is trans-abdominal rumen aspiration through the left paralumbar fossa, a technique that requires no preparation time, less restraint, and no special equipment. This method has been used for over 20 years by the primary investigator (JRR), but safety and efficiency have never been evaluated. The primary aim of this study was to determine if trans-abdominal ruminal aspiration is a quick, safe, and efficient method to collect rumen fluid to evaluate rumen pH and rumen protozoa.

Materials and Methods

The primary objective of this study was to document the methodology, determine the actual time of obtaining a rumen fluid sample, and to evaluate any complications when collecting rumen fluid by rumenocentesis through the left paralumbar fossa. A secondary objective was to determine rumen pH and the typical number of protozoa per 40x field in cows grazing a unique diet. Fifty-eight mature cows from the Ross University School of Veterinary Medicine teaching herd were used under an institution-approved animal care and use protocol. Cows used in the study were predominately Senepol and Senepol-cross breeds. These cattle are primarily fed guinea grass in the morning, with occasional supplementation with brewer's grain.

Procedures

Each cow was restrained in a traditional squeeze-chute; there was no skin preparation before the procedure. Prior to needle insertion, the left paralumbar fossa was evaluated for body wall thickness via ultrasonography, and measurements were recorded. Tail restraint was then applied, and a 1.5 inch (3.8 cm) 16-gauge needle attached to a 12-ml syringe was directed toward the right elbow in the lower "V" of the left paralumbar fossa (Figures 1 and 2). As soon as the needle was introduced through the skin, suction was applied to the syringe. The needle and syringe were pushed up to the hub of the needle, then the needle was withdrawn. Collection of 1 to 2 drops of rumen fluid was considered successful as both rumen pH and rumen protozoa can be evaluated with 1 drop. Immediately following collection, rumen pH was measured using pH paper,[a] and the pH was recorded for each cow. Rumen protozoa were evaluated using light-field microscopy at 40x magnification. A drop of rumen fluid was placed on a clean slide, and 3 fields were assessed for number of protozoa. An average number of protozoa from the 3 fields was calculated and recorded for each cow. A new needle and syringe were used on each animal. If the first attempt was unsuccessful, 1 additional attempt was performed. A stopwatch was used to time from insertion to extraction. After sample collection, the animal was released back into the herd.

Post-procedure

Cows were visually evaluated within 1 hour following the procedure, looking for any evidence of pain or swelling (day 0). Cows were then evaluated on days 1, 4, and 18 for any evidence of pathology of the paralumbar fossa via visual inspection and ultrasonography with body wall thickness measurements taken on each observation day. Evidence of possible pathology noted during ultrasonography were subjectively assessed as: none - absolutely nothing suggestive of pathology; minor - possible subtle lesions observed < 5 mm; moderate - external swelling with possible lesion observed, > 5 mm thickness from surrounding tissue; or major - obvious external swelling and definitive area of fluid density with increased body wall thickness seen within the body wall. Scoring was based on both visual assessment and ultrasonographic assessment. The ultrasonographic assessment was based on increased body wall thickness with possible pockets of pathology, such as hematoma or abscess.

Although day 18 represented the last structured evaluation of the cows in the study, the cows were used for teaching purposes throughout the year (physical examination with rumen assessment), and any obvious swellings in the paralumbar fossa would have been noticed.

Statistics

Simple descriptive statistics[b] were used for pH and rumen protozoal counts. A logistic regression model[c] was fit

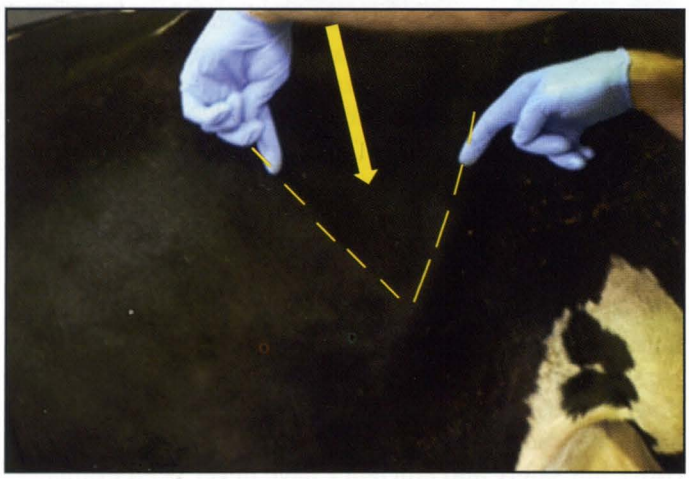

Figure 1. The "V" in the left paralumbar fossa. The yellow arrow indicates the site for the "tap", but anywhere in this general area where rumen can be palpated should suffice. The yellow hatched line represents the "V".

Figure 2. Needle should be inserted into the lower area of the "V". Yellow hatched line represents the "V" of the paralumbar fossa.

to the data to determine whether body wall thickness was predictive of rumen tap success at day 0. A simple logistic regression model was fit to the data ($y = b_0 + b_1x + \varepsilon$), where:

y is an indicator variable of rumen tap success (y=1) at day 0, y=0 otherwise;

b_0 is an intercept common to all cows;

b_1 is the regression coefficient pertaining to body wall thickness;

x is the body wall thickness (in millimeters); and

ε is an error term.

Results

Sufficient rumen fluid was collected for analysis from 78% of the 58 cows. Average body wall thickness for cows successfully tapped was 25.4 mm (range of 14 to 41 mm), whereas the average body wall thickness for cows not successfully tapped was 37.8 mm (range of 22 to 47 mm). A logistic regression model was fit to the data to determine if body wall thickness was predictive of rumen tap at day 0. Body wall thickness was a statistically significant ($P = 0.0002$) predictor of a successful rumen tap. Odds of successful rumenocentesis was >90% when the body wall thickness was < 20 mm, 62% when < 35 mm, and 36% when ≥ 40 mm.

The average time for successful rumenocentesis was 2.9 seconds, with a range of < 1 to 8 seconds. The median and mode times were 2.45 and 2 seconds, respectively. Average pH of evaluated rumen fluid was 7.9 with a range of 7 to 8.5, while the average number of mobile protozoa identified at 40x power was 3 (range 0 to 7), with an average maximum of 4 (range 0 to 9).

No externally visual pathology was noted on any cow during the study. Increased body wall thickness measurements > 5mm were present, based on ultrasonographic follow-up, for 11 of the 58 head (19%). The single case of moderate pathology, first seen on day 1, was thought to be a forming hematoma, but it did not increase in size or body wall thickness over the course of the study. Possible pathology based on ultrasonography, rather than body wall thickness, was compared to successful vs unsuccessful rumen fluid collection (Table 1).

Table 1. Possible pathology (hematoma/abscess development) as observed via ultrasonography.

Possible lesion*	All cows	Successful taps	Unsuccessful taps
None	55% (32/58)	53% (24/45)	62% (8/13)
Minor	43% (25/58)	44% (20/45)	38% (5/13)
Moderate	2% (1/58)	2% (1/45)	0% (0/13)

*None = no lesions observed; minor = possible subtle lesions observed; moderate = apparent lesion observed

Discussion

Paralumbar fossa trans-abdominal rumen fluid sampling in adult cattle was determined to be quick and safe; on average sampling took 3 seconds. The rapid collection time was in part because skin preparation is not necessary for the procedure. No other studies were found that reported the time required for rumen fluid collection. It is important to note, however, that this method is not a good alternative to that described by Nordlund and Garrett for assessment of sub-acute ruminal acidosis, as the volume of rumen fluid needed is higher than that easily achievable by the paralumbar fossa trans-abdominal approach.[4] The current described method is best for a quick assessment of ruminants suspected of acute ruminal acidosis or urea toxicosis (nonprotein nitrogen) and/or to assess the viability of ruminal protozoa to determine if rumen transfaunation is needed.

The average ruminal pH in the current study was higher than most reference ranges for cattle. Sampling was conducted prior to the morning feeding of guinea grass, which may be 1 reason that pH was higher as the cattle were not fed for ~24 hours prior to collection of rumen fluid.

Post-procedure pathology, such a hematoma, seroma, abscess, or peritonitis, was not observed in the study. No external evidence of pathology was seen in the study follow-up nor in any period following the procedure (Table 1). We anticipated having more pathology associated with unsuccessful aspirations due to the increased trauma of repeated efforts, but this was not the case. These cattle are used for teaching purposes throughout the year, and there was no evidence of long-term pathology at the puncture sites. Mialon and co-workers, who utilized a similar rumenocentesis site but also prepped the site (shaved and disinfected with an iodine solution), also did not report evidence of inflammation at the puncture site.[3] They further concluded that local anesthesia provided no welfare benefit or that any benefit was too small for detection by their assessment method. However, others have observed minor post-rumenocentesis pathology. Nordlund and Garrett, who utilized a ventral rumen sac approach, reported up to 2% abscesses at the puncture site.[4] Noro et al, who performed a dorsomedial rumenocentesis technique every 5 days for 1 month, reported temporary swelling (0.4 to 0.6 in; 1 to 1.5 cm) at the puncture site 4% of the time.[5] Tajik et al utilized a method similar to Nordlund and Garrett, and reported a small local reaction in 23 of 196 (11.7%) cows, and a single cow developed a small superficial abscess that cured spontaneously.[7] No other general health impairment was observed. Using ultrasonography, we identified possible pathology in 45% of 58 head, but no grossly observable pathology. In a study by Kleen and others on 164 dairy cows, 9 head (5.5%) of the study population showed alterations at the puncture site, such as hematoma or abscess formation, and the general health status was compromised in 3 cows after collection.[2] Yet, the authors considered rumenocentesis a viable diagnostic procedure in bovine health diagnostics. Strabel and others, using a ventral rumen sac sampling site,

reported severe complications following rumenocentesis as 9 of 11 (82%) of cows developed hematoma formation, and 1 cow developed severe generalized septic peritonitis.[6] Rumen fluid collection in the present study was performed without skin preparation, and no complications were observed.

The success of our procedure for obtaining rumen fluid was 78%. Increased efficiency in obtaining rumen fluid could be achieved by using a longer needle (e.g 3 inch or 7.6 cm) for cows with a high body condition score. To our knowledge, this is the first report of rumenocentesis utilizing a 1.5 inch (3.8 cm) 16-gauge needle. This technique allows a practitioner to successfully obtain a rumen sample in order to effectively diagnose and treat ruminant patients in a quick, safe, and efficient manner without the need for specialized equipment.

Conclusion

Under the conditions of this study, the rumenocentesis technique described here is quick, simple, and safe, and is most efficient for cattle with a relatively thin body wall.

Endnotes

[a]pHydrion® pH Paper, Micro Essential Laboratory, Brooklyn, NY

[b]Microsoft Excel 2010, Redmond, WA

[c]R, Vienna, Austria

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