PEER REVIEWED

Growth Performance of Cattle Following Percutaneous Liver Biopsy Utilizing a Schackelford-Courtney Biopsy Instrument

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Abstract

Percutaneous liver biopsy has been utilized routinely in cattle as a diagnostic and research tool. However, there is little information on the effect on post-procedural performance. This paper describes a modified technique utilizing a Schackelford-Courtney bovine liver biopsy instrument, which generally obviates the need for multiple entries.

Two trials were performed to evaluate post-procedural growth performance. In Trial 1, performed on finishing cattle, average daily gain (ADG) from the date the procedures (liver biopsy, ruminocentesis, orogastric intubation) were performed through the end of the 120-day finishing period was not different (p>0.25). However, calves that underwent the procedures had lower ADG, dry matter intake (DMI) and gain-to-feed ratio (G:F) (p<0.01) at Day 7 than calves that did not undergo the procedures. In Trial 2, a 112-day pasture study using stocker heifers, there were no significant differences at any weigh day (7, 28, 56, 84, 112) between liver-biopsied and non-biopsied groups. Results of the second trial suggest that differences seen in Trial 1 at Day 7 could have been related to other procedures performed concurrently (ruminocentesis, orogastric intubation), rather than the liver biopsy itself. In both studies, percutaneous liver biopsy was a relatively safe and effective procedure with minimal complications.

Résumé

La biopsie transcutanée du foie est utilisée de façon routinière chez le bétail comme outil de diagnostic et de recherche mais il y a peu d'information sur la performance suivant la procédure. Cet article décrit une technique modifiée qui repose sur l'instrument de biopsie du foie bovin de Schackelford-Courtney qui réduit généralement le besoin de faire des entrées multiples.

Deux essais ont été conduits pour évaluer la performance de croissance suite à la procédure. Dans l'essai 1, mené avec du bétail en engraissement, le gain moyen quotidien (GMQ) mesuré depuis la date des procédures (biopsie du foie, rumenocentèse, intubation orogastrique) jusqu'à la fin de l'engraissement 120 jours plus tard n'était pas différent (p > 0.25). Toutefois, les veaux qui subirent la procédure avait un GMQ plus bas, une ingestion de matière sèche et une conversion alimentaire plus faibles (p < 0.01) au jour 7 que les veaux qui n'avaient pas subi les procédures. Dans l'essai 2, une étude au pâturage de 112 jours avec des taures à l'engraissement, il n'y avait pas de différence à chacune des périodes de prise du poids (7, 28, 56, 84, 112) entre les groupes ayant subi ou non la biopsie du foie. Les résultats de l'essai 2 suggèrent que les différences lors de l'essai 1 auraient pu être causées par les autres procédures faites au même moment (rumenocentèse, intubation orogastrique) plutôt que par la biopsie du

foie elle même. Pour chaque essai, la biopsie transcutanée du foie était une procédure relativement sécuritaire et effective avec très peu de complications.

Introduction

Liver biopsies were first performed in human patients in 1833,[11] and a liver biopsy technique in sheep was described by Dick in 1944.[7] Several techniques for this procedure have been described, with protocols including a highly invasive partial lobectomy, guided approaches using ultrasound or a laparoscope, and blind approaches.[4,6,7,11,12,14,16,17] Reasons for collecting samples include diagnostic biopsies in a hospital environment, and field use for herd studies with a representative number of animals. Percutaneous liver biopsy has been used for many years in cattle to diagnose hepatic disease syndromes and to evaluate trace mineral status. This paper describes an efficient field liver biopsy technique and an evaluation of the growth performance of calves following the biopsy. Both feeding and pasture trials were performed to evaluate growth performance in different management systems.

There has been some reluctance by practitioners to perform routine field liver biopsies in cattle due to potential problems with mortality and growth performance.[11] One report states that liver biopsy should only be done after checking prothrombin or whole blood clotting time.[13] Reported contraindications to performing a liver biopsy include peritonitis, abscessation, biliary obstruction and the suspected presence of vascular tumors.[13] Nonetheless, reports of complications from the procedure have been rare.[11,13] The Tru-Cut biopsy needle[a] has proven to be a reliable and safe instrument for collection of liver biopsy specimens.[1,5,8,9,13] However, retrieval of enough tissue for trace mineral analysis usually requires three biopsy entries with this instrument. When using the Schackelford-Courtney instrument and a modified technique described in this paper, at least 150 mg of liver tissue (wet weight) per biopsy is typically harvested, thus minimizing the need for multiple sampling to obtain sufficient tissue for trace mineral analysis.

Instrumentation and Biopsy Technique

The Schackelford-Courtney bovine liver biopsy instrument[b] was used in these studies. This instrument consists of a side-notched obturator sheathed by a 5-mm diameter needle (Figure 1). The animal is restrained in a squeeze chute, and the point of insertion for the biopsy instrument is located. The point of insertion of the instrument is in the right 10[th] intercostal space along an imaginary line drawn from the top of the tuber coxae to the point of the right shoulder (Figure 2). The 10[th] intercostal space is best identified by locating the 13[th] rib and counting cranially, beginning with the 12[th] intercostal space. A 10 x 10 cm area, centered over the point of insertion, is clipped and prepped with a povidone-iodine scrub and disinfectant solution. Anesthesia is provided by the local infiltration of 3 to 5 ml of 2% lidocaine, followed by final surgical preparation of the site.

A 1-cm stab incision is made through the skin at the point of insertion. The biopsy instrument is passed through the skin incision and directed at an angle toward the opposite elbow (Figure 2). The instrument is

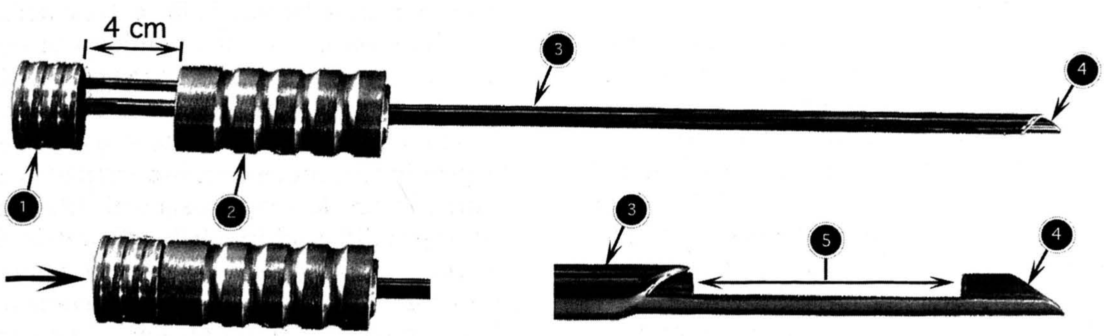

Figure 1. Photograph of biopsy instrument with obturator sheathed for insertion and withdrawal (upper) and with obturator extended to expose specimen notch (lower):
1) obturator control handle;
2) needle control handle;
3) needle;
4) obturator;
5) specimen notch.

[a]Allegiance Healthcare Corporation, McGaw Park, IL 60085
[b]Schackelford-Courtney Bovine Liver Biopsy Instrument, Sontec Instruments, Englewood, CO 80112

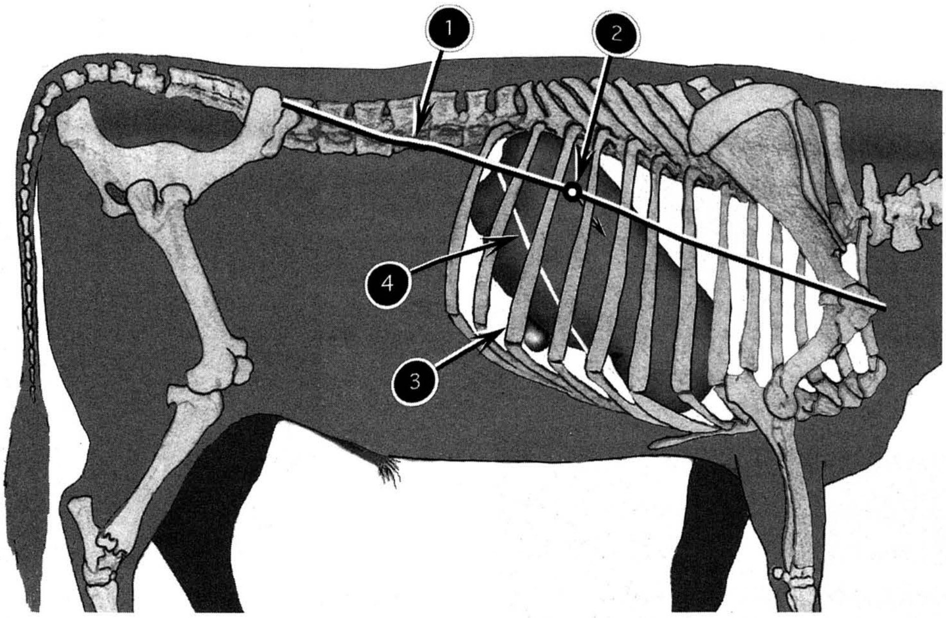

Figure 2. Anatomical landmarks for bovine liver biopsy technique:
1) line from the top of the right tuber coxae, to the point of the shoulder;
2) point of insertion of biopsy instrument along line in 10th intercostal space; instrument is directed cranioventrally toward the opposite elbow;
3) 11th rib;
4) approximate line of pleural reflection.

passed through the diaphragm and into the liver with the obturator in the withdrawn position (Figure 3A). Once in the liver, the outer needle is held steady and the obturator is advanced to expose the specimen notch (Figure 3B). The obturator handle is then held steady and the needle is advanced to "resheath" the obturator and capture the biopsy specimen (Figures 3C and 4). With the instrument maintained in this position, the device is withdrawn, and the skin incision is sutured using a single cruciate pattern with #1 Vetafil suture. Other suture materials could be used according to individual preference. Prophylactic penicillin should be routinely administered in areas where Redwater disease (*Clostridium hemolyticum*) is a problem. Prophylactic penicillin or other antimicrobials were not administered in the studies described.

Approximately 150 mg (wet weight) of liver tissue is consistently obtained with this technique. The minimum recommended sample for a complete trace mineral analysis by inductively coupled plasma-atomic emission spectroscopy with ultrasonic nebulization (ICP) is 50 mg (wet weight).[2,3]

Materials and Methods

Trial 1 – Evaluation of steer finishing growth performance following percutaneous liver biopsy and ruminal fluid collection by two methods

Sixty steers (average weight 871.2 lb; 396 kg) were blocked by weight and placed in five pens (12 steers/pen). Calves were used in an 84-day backgrounding trial assessing growth performance when fed recycled poultry bedding-based diets. At the end of the 84-day trial, six animals from each pen were randomly selected for sample collection procedures, which included liver biopsy, ruminal fluid collection by ruminocentesis and orogastric intubation. Randomization was done by drawing numbered beads from a hat. The ruminocentesis method described by Nordlund and Garrett[10] was used to avoid salivary contamination of ruminal fluid. Ruminal fluid was also collected by manual passage of a tube through the oral cavity and into the rumen. Ruminal fluid collection by ruminocentesis and orogastric intubation was performed for comparative analysis and will not be further discussed in this paper. A percutaneous liver biopsy (utilizing the technique described above) was obtained to assess trace mineral status. Generally, only one attempt was necessary to obtain an adequate sample. A total of three entries were performed before considering the procedure unsuccessful.

Calves were maintained on study diets for one additional week after samples were collected. Calves were then placed on a 2-week transition diet schedule to acclimate them to higher-grain finishing diets. The finishing period was approximately 120 days. All calves were fed the same transition and finishing period diets.

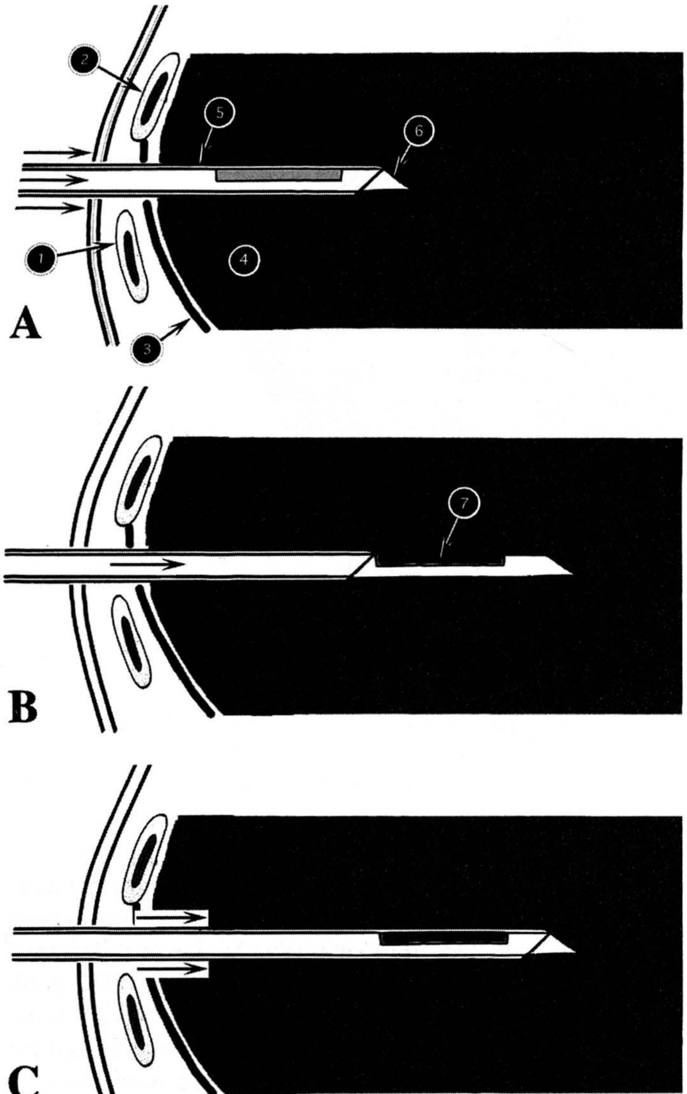

Figure 3. Schematic representation of percutaneous liver biopsy technique:
1) 10th rib;
2) 11th rib;
3) diaphragm;
4) right lobe of liver;
5) needle;
6) obturator;
7) specimen notch;
A) initial insertion of instrument with obturator in withdrawn position;
B) needle handle held steady and obturator advanced to expose specimen notch;
C) obturator handle held steady and needle advanced to capture biopsy specimen prior to withdrawal of instrument.

Figure 4. Photograph of biopsy instrument fully inserted in right 10th intercostal space. At this point, the obturator has already been advanced beyond the needle. The obturator handle is being held steady, while the needle handle is being advanced to cover the specimen notch prior to withdrawal of the instrument (see Figure 3C).

Calves were fed in individual feeders[c] and dry matter intake (DMI) was monitored throughout the backgrounding, transition and finishing periods. Calf weights were recorded on the two days prior to the procedures, one week post-procedures and at the end of the transition and finishing periods. Gain-to-feed (G:F) ratios were calculated from individual calf average daily gain (ADG) and DMI. Differences were assessed in ADG, DMI and G:F between calves that underwent procedures and those that did not using SAS® general linear models procedure.[15] Effects of procedures and pen were determined. Since two technicians performed the percutaneous liver biopsy procedure in this trial, technician effect was also determined.

Trial 2 – Evaluation of growth performance of stocker heifers following percutaneous liver biopsy

Sixty-six crossbred heifers (average weight 563 lb; 256 kg) were purchased from North Carolina state graded feeder calf sales in April 2000. Following a six-week backgrounding period, heifers were ranked by weight and randomly allotted from within weightblocks to one of six treatments. Randomization was performed by drawing numbered beads from a hat.

Treatments were arranged in a 2 x 3 factorial design with the factors being liver biopsy or no liver biopsy, and one of three implant regimes: no implant; 40 mg trenbalone acetate - 8 mg estradiol;[d] or 36 mg of zeranol.[e] Calves were weighed during the morning on

[c]Calan Gates, American Calan, Inc., Northwood, NH 03261
[d]Revalor®-G, Hoechst Roussel Vet, Warren, NJ 07059
[e]Ralgro®, Schering-Plough Animal Health, Union, NJ 07083

two consecutive days at the beginning of the trial. Following the second weighing, liver biopsies were performed and growth implants were administered. Cattle were re-weighed during the morning on Days 7, 28, 56, 84, 111, and 112. During each weighing, the biopsy instrument insertion site was carefully observed for complications. Cattle were managed as one group, and were rotationally grazed on mountain pastures consisting primarily of endophyte-infected KY-31 tall fescue, orchardgrass, bluegrass, and red and white clovers. Cattle had access to a free-choice mineral and fresh water at all times.

Results and Discussion

Steers in Trial 1 were well restrained and relatively calm during all invasive procedures. The biopsy attempt was successful in 30 of 31 calves. Liver tissue was not obtained from one calf after three attempts; therefore, the procedure was discontinued. In a second calf, a slight amount of intestinal content was observed after the first attempt, but liver tissue was obtained during the second attempt. Another calf developed a subcutaneous abscess at the biopsy site and exhibited anorexia. This steer recovered uneventfully following drainage of the abscess and parenteral treatment with procaine penicillin G. The suture material utilized may have contributed to the formation of the subcutaneous abscess. This steer was included in the data analysis because it represented a potential complication of percutaneous liver biopsy.

In Trial 1, the ADG of steers from the date procedures were performed through the finishing period was not different ($p>0.25$; Table 1). However, calves that underwent sample collection procedures had lower ADG, DMI and G:F ($p<0.01$) during the first week post-procedures than calves that did not undergo the procedures. The calves were able to compensate, as evidenced by their performance in the transition and finishing periods. Performance in the finishing period was not affected by the sample collection procedures ($p>0.30$).

Pen effects in Trial 1 were significant ($p<0.02$) for DMI and G:F during the finishing period, as well as ADG and G:F during the period immediately prior to the procedures. This was a direct result of blocking the animals by weight within a pen. Lighter animals would presumably be in a steeper phase of their growth curve and gaining more weight while consuming less feed, leading to more efficient gain. Because pen effects were significant, they were left in the model. There was no interaction between pen and sample collection procedures ($p>0.20$). Technician effect was not significant ($p>0.10$).

Trial 1 ADG, DMI and G:F were measured in the same animals over a period of time. Therefore, the measures were not independent of one another. A repeated measures multivariate analysis of ADG was also performed. The results were the same as those found using a simple analysis of variance. The sample collection procedures had a significant effect on ADG during week one after the procedures were performed, but animals

Table 1. Growth performance of steers following percutaneous liver biopsy, rumenocentesis, and orogastric intubation in Trial 1.

Parameter	Underwent procedures	Did not undergo procedures	p-value*
Number of animals	31	29	N/A
Start weight lb (kg)	865 (393)	880 (400)	0.13
7-d post procedures			
Average daily gain lb (kg)	0.44 (0.20)	2.84 (1.29)	0.0014
Dry matter intake lb (kg)	20.72 (9.42)	23.52 (10.69)	0.0058
Feed efficiency (G:F)	0.003	0.12	0.0029
Transition period			
Average daily gain lb (kg)	3.41 (1.55)	3.32 (1.51)	0.76
Dry matter intake lb (kg)	19.01 (8.64)	19.58 (8.90)	0.16
Feed efficiency (G:F)	0.18	0.17	0.44
Finishing period			
Average daily gain lb (kg)	2.57 (1.17)	2.57 (1.17)	0.92
Dry matter intake lb (kg)	18.19 (8.27)	18.72 (8.51)	0.35
Feed efficiency (G:F)	0.14	0.14	0.38
Total average daily gain lb (kg)	2.53 (1.15)	2.64 (1.20)	0.30
Final weight lb (kg)	1195 (543)	1214 (552)	0.11

*p-value is level of significance of difference in average values by treatment.

compensated and there was no difference in ADG during the transition and finishing periods.

At the end of the finishing period, carcass traits were collected at harvest and analyzed by procedures and pen. There was no interaction between pen and performance of sample collection procedures (p>0.05), and no effect of pen (p>0.10), so only the effect of the sample collection procedures was used in the final model. There was no difference in marbling, quality grade or fat thickness between calves that underwent the procedures and those that did not (p>0.30; Table 2). Ribeye area tended to be greater for calves that did not undergo the procedures than for those that did (p<0.09). This can be explained by the slightly lower hot carcass weight of the calves which had been sampled. No differences occurred in ribeye area per lb (kg) of carcass weight. Hot carcass weight and dressing percent were lower for calves undergoing the sampling procedures (p<0.05). This difference was a function of slightly lower final live weight of calves that underwent the procedures (p>0.10; Table 1). The slightly lower final live weight of calves that underwent sampling procedures was a direct result of an insignificant difference in body weight prior to the procedures (p>0.10; Table 1). Calves were randomly selected for procedures, so the difference in body weight was a chance event.

In Trial 2, adequate liver samples were collected from 26 of 33 heifers for ICP trace mineral analysis (> 10 mg dry weight or > 50 mg wet weight).[13] The procedure yielded liver biopsy samples with an average weight of 194.5 ± .06 mg on a wet basis and 47.2 ± .01 mg on a dry basis. In most cases (23/33), a single entry was sufficient to obtain an adequate sample for analysis. After three unsuccessful entries, no further attempts were made to obtain a sample due to potential for unnecessary stress and trauma. In three heifers, we were unable to harvest a liver sample after three attempts. The fractious nature of these heifers increased the difficulty of the procedure. A slight amount of intestinal content was observed after one attempt in one calf, and another calf exhibited severely altered mentation, resulting in two unsuccessful attempts. Biopsy was attempted on all calves that were prepared for biopsy (33), therefore 33 heifers were included in the biopsy treatment group for analysis.

On Day 7, a small swelling (18-23 mm in diameter) was noted in 12 of the 33 biopsied heifers. By Day 28, observable swelling had disappeared from all heifers, and no further complications were noted during subsequent weigh periods.

Weight block effects were not significant for any variable in Trial 2, so block was removed from the final statistical model. There were no significant differences between treatments at any weigh period during this trial (Table 3). Four heifers were removed from the trial for reasons unrelated to treatment:

Table 2. Carcass characteristics of steers following percutaneous liver biopsy, rumenocentesis, and orogastric intubation in Trial 1.

Parameter	Underwent procedures	Did not undergo procedures	p-value[1]
Number of animals	31	29	N/A
Marbling[2]	5.63	5.38	0.30
Quality grade[3]	17.0	16.7	0.39
Ribeye area (sq in)	12.66	13.13	0.08
Back fat (in)	0.49	0.46	0.42
Hot carcass wt lb (kg)	713 (324)	737 (335)	0.03
Dressing percent	59.58	60.30	0.03

[1]p-value is level of significance of difference in average values by treatment.
[2]Marbling: 4 = slight
5 = small
6 = modest
7 = moderate
[3]Quality Grade: 20 = prime –
19 = Choice +
18 = Choice
17 = Choice –
16 = Select +
15 = Select

Table 3. Effect of liver biopsy on gain of stocker heifers*.

Measurement	Biopsy	No biopsy	SEM**
Starting wt., lb (kg)	563.2 (256.0)	563.1 (255.9)	4.9
7-d ADG, lb (kg)/d	4.25 (1.93)	4.75 (2.16)	0.143
28-d ADG, lb (kg)/d	3.37 (1.53)	3.48 (1.58)	0.052
84-d ADG, lb (kg)/d	1.85 (0.841)	1.92 (0.873)	0.028
126-d ADG, lb (kg)/d	1.74 (0.791)	1.79 (0.814)	0.022
Final wt., lb (kg)	783.0 (355.9)	788.7 (358.5)	6.32

*No significant differences were observed.
**SEM calculations performed using kilograms of weight or weight change.

1) undetermined neurologic mortality (no biopsy group);
2) limb fracture;
3) extremely wild and could not be weighed on all dates; and
4) parturition.

The lack of difference in ADG between treatments on Day 7 was surprising, and suggests that post-procedural feed intake was not depressed as observed in Trial 1. Furthermore, these heifers were quite excitable and represent a practical situation in which complications might be expected, which makes the lack of treatment effect even more intriguing. The difference in ADG on Day 7 in Trial 1 between the biopsy and no-biopsy treatment groups may have been the result of other procedures (ruminocentesis, orogastric intubation), which may have confounded the effect of biopsy on ADG and may have had substantial negative impact on the ADG during the first week. Anecdotal observations at this research farm of decreased intake after orogastric intubation offers some justification to this conclusion. Also, the technician performing the liver biopsy procedure in trial 2 (GR) was one of the technicians in Trial 1; therefore he had more experience with the Schackelford-Courtney instrument prior to the start of Trial 2.

Several other reasons could explain the results of Trial 2 compared to Trial 1. Since this was a grazing study as compared to a feeding study, one would expect the gain differences to be smaller when gains are lower. In Trial 1, two technicians conducted biopsy procedures, as compared to one technician in Trial 2. However, no differences were seen in Trial 1 when the data was analyzed by technician effect. The technician performing the procedure in Trial 2 (GMR) had used this procedure on several other occasions since Trial 1. This may indicate the positive effect of experience conducting liver biopsies. Most importantly, the calves in Trial 1 were exposed to ruminocentesis and oral rumen fluid collection at the same time that percutaneous liver biopsy was performed. Despite the performance of all three sample collection procedures in Trial 1, differences in growth performance were only seen at the 7-day weigh period.

In these trials, significant detrimental effects on growth performance following liver biopsy were not detected, given the power of the statistical test utilized. In Trial 1, there was sufficient power to detect an overall ADG difference of 0.33 lb (0.15 kg)/d; in Trial 2, we could detect a difference in overall ADG of 0.22 lb (0.10 kg)/d. The results of these studies demonstrate, with a reasonable level of power, that an effect of liver biopsy on growth performance was either non-existent or minimal. It cannot be accurately stated that there was no effect, but rather that no effect could be identified given the power of the statistical test.

Conclusions

We conclude that routine liver biopsy of a representative sample size (*e.g.* 10% of herd) in a field environment should be considered when herd diagnostic challenges (such as suspected copper deficiency, copper toxicity, or pyrrolizidine alkoloid toxicity) occur.

These trials demonstrated that a reduction in post-procedure growth performance was minimal after percutaneous liver biopsy using the technique described. The Schackleford-Courtney bovine biopsy instrument was relatively safe and effective for collection of liver samples in these trials, which confirms previous statements by other authors about the relative safety of liver biopsy. To our knowledge, this is the first report providing quantitative evidence of the minimal impact of a field liver biopsy procedure on growth performance.

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Abstracts

Septicaemia in a Pig Farm Worker

E. J. Watkins, P. Brooksby, M. S. Schweiger, S. M. Enright
Veterinary Record (2001):148, 629

The death of a 30-year-old pig farm worker from *Streptococcus suis* type 14 sepsis is reported. The organism was identified as the cause of a lameness outbreak in pigs which was under investigation at the time of the farmworker's illness. His main work was to paint the outside of the pig sheds and once weekly to pressure-hose the weaning pens. It is possible that infection entered through a foot abrasion as a result of contaminated water soaking into his boot. Alternatively he may have inhaled contaminated water spray or transferred bacteria from his hands to mouth. The patient had undergone a splenectomy three years previously following trauma.

Restocking of a Dairy Farm

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Veterinary Record (2001):148, 623

Following depopulation due to the confirmation of a BSE-positive cow, the Moorepark research centre farm in Ireland was restocked with 297 pregnant heifers and 105 weaner heifers registered with the Irish Holstein Fresian Society. Prepurchase tests for tuberculosis (TB), brucellosis, *Mycoplasma bovis* and Johne's disease were performed. The antibody status to bovine viral diarrhoea (BVD) virus and infectious bovine rhinotracheitis (IBR) was also determined in the animals to be purchased. On arrival all animals were given an anthelmintic and injected with streptomycin/dihydrostreptomycin, vaccinated for leptospirosis and IBR, and were maintained in separate groups according to source. Of three animals which aborted two tested negative for brucellosis and one was slaughtered as a precaution. The fetuses did not provide a definitive diagnosis. Six cows were culled postpartum for: limb fracture (three), tarsitis (one), bloat (one) and abortion (one). Of 32 prenatal deaths, 30 were stillbirths. A positive diagnosis was made in eight calves for: thyroid hyperplasia (four), anoxia (one), amniotic fluid in bronchi (one), neosporosis (one) and high nitrite concentration (one). The costs of the tests were thought to be so high as to be neither practical nor economical for an individual farmer, but nevertheless someone intending to purchase similar animals should test not only for TB and brucellosis, but also, prepurchase, for mycoplasma, BVD (immunoper-oxidase test), and Johne's disease. Herd health status was maintained for the three years following purchase in this complex biosecurity exercise.