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EVALUATION OF THE LAPAROSCOPIC LIVER BIOPSY TECHNIQUE WITH BABCOCK FORCEPS IN CADAVERS AND CANINE PATIENTS

AVALIAÇÃO DA TÉCNICA DE BIÓPSIA HEPÁTICA LAPAROSCÓPICA COM PINÇA BABCOCK EM CADÁVERES E PACIENTES CANINOS

EVALUACIÓN DE LA TÉCNICA DE BIOPSIA HEPÁTICA LAPAROSCÓPICA CON FUERZAS DE BABCOCK EN CADÁVERES Y PACIENTES CANINOS

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ABSTRACT

Considering that laparoscopic liver biopsy is a diagnostic technique but often relies on specialized equipment, evaluating accessible alternatives is necessary. The objective was to evaluate the feasibility and efficiency of a laparoscopic liver biopsy technique using Babcock forceps. To this end, we proceeded to a two-phase study: an initial ex vivo phase comparing Babcock and conventional forceps in a bovine fetus model, followed by an in vivo phase in six canine patients with suspected liver disease. We assessed procedural time, sample quality (area, crushing artifacts, number of portal spaces), and complications. Thus, it is observed that the technique was rapid (mean 13.2 minutes) and no intraoperative hemorrhage occurred. The samples were sufficient for the production of histopathological sections and diagnosis, with a crushing artifact of 21.62% that did not preclude a definitive histopathological diagnosis for conditions including neoplasia, cholangitis, and chronic hepatopathies. This allows us to conclude that the laparoscopic liver biopsy technique using Babcock forceps is a feasible, safe, and efficient method, being a viable alternative for obtaining liver samples in dogs.

Keywords: Laparoscopy. Liver Biopsy. Babcock Forceps. Dogs. Minimally Invasive Surgery.

RESUMO

Considerando que a biópsia hepática laparoscópica é uma técnica diagnóstica, mas frequentemente depende de equipamentos especializados, é necessário avaliar alternativas

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Revista ARACÊ
ISSN: 2358-2472

acessíveis. O objetivo foi avaliar a viabilidade e a eficiência de uma técnica de biópsia hepática laparoscópica usando pinça Babcock. Para tanto, procedemos a um estudo de duas fases: uma fase inicial ex vivo comparando pinça Babcock e convencional em um modelo de feto bovino, seguida por uma fase in vivo em seis pacientes caninos com suspeita de doença hepática. Avaliamos o tempo do procedimento, a qualidade da amostra (área, artefatos de esmagamento, número de espaços portais) e as complicações. Assim, observa-se que a técnica foi rápida (média de 13,2 minutos) e não ocorreu hemorragia intraoperatória. As amostras foram suficientes para a produção de cortes histopatológicos e diagnóstico, com um artefato de esmagamento de 21,62% que não impediu um diagnóstico histopatológico definitivo para condições como neoplasia, colangite e hepatopatias crônicas. Isso nos permite concluir que a técnica de biópsia hepática laparoscópica com pinça Babcock é um método factível, seguro e eficiente, sendo uma alternativa viável para obtenção de amostras de fígado em cães.

Palavras-chave: Laparoscopia. Biópsia Hepática. Pinça Babcock. Cães. Cirurgia Minimamente Invasiva.

RESUMEN
Considerando que la biopsia hepática laparoscópica es una técnica diagnóstica pero a menudo depende de equipo especializado, evaluar alternativas accesibles es necesario. El objetivo fue evaluar la factibilidad y eficiencia de una técnica de biopsia hepática laparoscópica utilizando pinzas Babcock. Para este fin, procedimos a un estudio de dos fases: una fase ex vivo inicial comparando pinzas Babcock y convencionales en un modelo de feto bovino, seguida de una fase in vivo en seis pacientes caninos con sospecha de enfermedad hepática. Evaluamos el tiempo de procedimiento, la calidad de la muestra (área, artefactos de aplastamiento, número de espacios portales) y las complicaciones. Por lo tanto, se observa que la técnica fue rápida (media de 13,2 minutos) y no se produjo hemorragia intraoperatoria. Las muestras fueron suficientes para la producción de cortes histopatológicos y el diagnóstico, con un artefacto de aplastamiento del 21,62% que no impidió un diagnóstico histopatológico definitivo para afecciones que incluyen neoplasia, colangitis y hepatopatías crónicas. Esto nos permite concluir que la técnica de biopsia hepática laparoscópica mediante pinza Babcock es un método factible, seguro y eficiente, siendo una alternativa viable para la obtención de muestras de hígado en perros.

Palabras clave: Laparoscopia. Biopsia Hepática. Pinzas Babcock. Perros. Cirugía Mínimamente Invasiva.

Revista ARACÊ
ISSN: 2358-2472

1 INTRODUCTION

High-quality liver biopsy remains a cornerstone for the definitive diagnosis and staging of hepatic disease in dogs, enabling the histopathological identification of persistent laboratory abnormalities and the evaluation of disease progression (KEMP et al., 2015). It is particularly indicated in cases of unexplained hepatomegaly, diffuse hypoechogenicity on ultrasound, focal nodules, and for monitoring therapeutic responses, establishing itself as a safe and effective diagnostic modality (MCDEVITT et al., 2016).

Minimally invasive techniques for obtaining liver samples are continually advancing. Laparoscopic liver biopsy (LLB) offers the significant advantage of direct visualization of the abdominal organs, allowing for targeted sampling of macroscopically affected areas, a benefit not afforded by blind percutaneous techniques (BALSA; CULP, 2019). Studies have demonstrated that laparoscopic samples are comparable in diagnostic quality to those obtained via celiotomy and superior to percutaneous needle biopsies (KEMP et al., 2015). Various instruments are employed in LLB, including Tru-Cut needles, cup biopsy forceps, and pre-tied ligatures, each with distinct advantages and limitations concerning sample size, artifact induction, and risk of hemorrhage (BUOTE; LOFTUS; MILLER, 2022; FERNANDEZ et al., 2017).

The Babcock forceps is a staple instrument in laparoscopic surgery, primarily used for the atraumatic grasping and manipulation of hollow organs and delicate tissues (SINGH; CASE, 2022). However, its application specifically for liver biopsy collection is novel and has not been formally evaluated in the veterinary literature. Investigating alternative instruments is of clinical relevance, as it can provide viable techniques for settings with limited access to specialized biopsy equipment. The initial evaluation of new surgical techniques in cadaveric models is a established practice, providing valuable data on feasibility, instrumentation, and procedural steps before clinical application (BUOTE; LOFTUS; MILLER, 2022; DOBBERSTEIN et al., 2024). While acknowledging the inherent limitations of cadaveric tissue, including post-mortem changes that may affect tissue integrity, such models serve as a crucial preliminary step for technique standardization and refinement.

Therefore, this study aimed to evaluate the feasibility and efficiency of a laparoscopic liver biopsy technique using Babcock forceps. The investigation was conducted in two phases: an initial *ex vivo* phase using a bovine fetus cadaveric model to compare the technique against conventional biopsy forceps, followed by an in vivo phase in canine patients with suspected liver disease. We assessed procedural times, sample quality

Revista ARACÊ
ISSN: 2358-2472

(including macroscopic and microscopic area, crushing artifacts, and number of portal spaces), and intraoperative complications to determine the technique's diagnostic utility and safety.

2 MATERIAL AND METHODS

O ethical approval and study design

This study was approved by the Animal Ethics and Welfare Committee of the Federal University of Pará (CEUA/UFPA, protocol no. 5028050717) and conducted in accordance with the guidelines of the National Council for Control of Animal Experimentation (CONCEA), Brazil. A two-phase, prospective study was designed. The initial ex vivo phase utilized a bovine fetus cadaveric model to allow for the direct comparison of the novel technique against a standard method in a controlled setting, enabling technique refinement and preliminary assessment of sample quality without patient risk (BUOTE; LOFTUS; MILLER, 2022). This was followed by an in vivo phase in canine patients to evaluate the clinical feasibility, safety, and diagnostic yield of the technique.

Ex vivo cadaveric phase

Six bovine fetus cadavers, with weights ranging from 20 to 30 kg, were sourced from a local inspected slaughterhouse. The use of a bovine fetus model provided organs of a scale comparable to medium-sized dogs and allowed for the collection of multiple paired samples. It is acknowledged that post-mortem changes, including tissue autolysis and the loss of vascular pressure and coagulation, limit the model's ability to predict clinical hemorrhage (DOBBERSTEIN et al., 2024). However, it remains a valid and established model for initial technique development and the comparative evaluation of sample size and gross architectural preservation (BUOTE; LOFTUS; MILLER, 2022; SINGH; CASE, 2022). The cadavers were kept frozen after death and thawed for 3 hours at room temperature prior to the procedures to standardize tissue consistency.

In vivo clinical phase

Six client-owned dogs with clinical and laboratory evidence of hepatobiliary disease, admitted to the Veterinary Hospital of the Federal University of Pará, were enrolled. Informed consent was obtained from all owners. Preoperative assessment included a complete blood count, serum biochemical profile, coagulation tests, and abdominal ultrasonography.

Laparoscopic liver biopsy procedure

All procedures were performed under general anesthesia. Dogs were positioned in

Revista ARACÊ
ISSN: 2358-2472

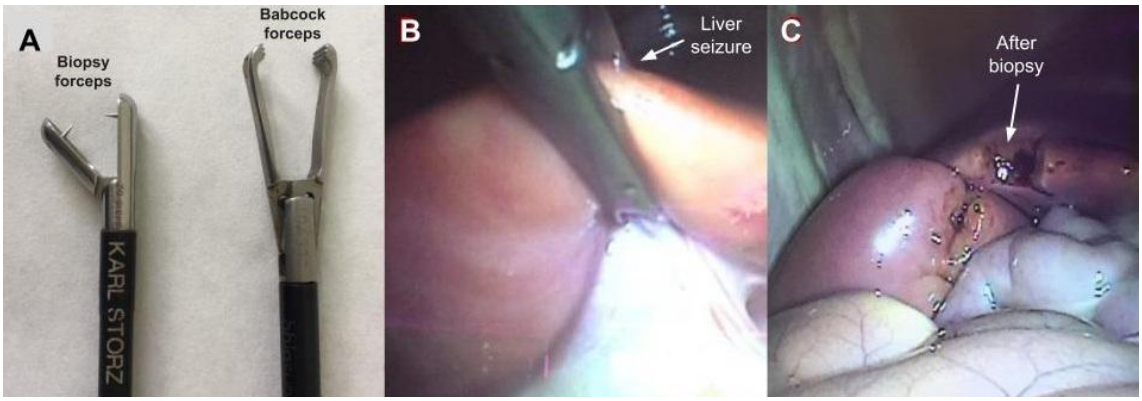
dorsal recumbency. A two-port technique was employed: a 10-mm optical trocar was placed on the ventral midline for a 30° laparoscope, and a 5-mm working port was established under direct visualization. Pneumoperitoneum was established and maintained with carbon dioxide insufflation at an intra-abdominal pressure of 8-10 mmHg.

The liver was systematically inspected. Biopsy sites were selected from lobes with macroscopic abnormalities (e.g., discoloration, nodularity). In the absence of discrete lesions, samples were taken from the edge of the left lateral or left medial liver lobes, as per standard practice (KEMP et al., 2015).

A 5-mm reusable Babcock forceps (Bf) characterized by its fenestrated, rounded jaws designed for atraumatic grasping, was used (Figure 1A). The forceps were used to grasp a fold of hepatic tissue for 5 seconds to induce localized ischemia. The tissue fold was then excised at its base using the sharp edge of the 5-mm trocar sheath, and the sample was retrieved through the instrument port. A 5-mm reusable cup biopsy forceps (Cf), with sharp, spoon-shaped jaws, was used as the control (Cf). The forceps were opened, pressed against the liver surface, closed, and withdrawn to retrieve a sample.

Figure 1

F*orceps and procedures of liver biopsies: (A) Image of laparoscopic forceps and procedures used in the study of liver biopsies, and extremities of the biopsy and Babcock forceps. Images of the technique with the Babcock forceps, (B) internal vision of liver seizure, and (C) internal vision immediately after biopsy.*



In the cadaveric phase, paired samples (Bf1 and Cf1, n=6 each) were collected from adjacent sites. In the clinical phase, 1 to 3 samples per dog were collected exclusively using the Babcock forceps, figure 1B, (Bf2, n=6). Each biopsy site was observed laparoscopically for a minimum of one minute for any signs of hemorrhage before trocar removal (Figure 1C).

Revista ARACÊ
ISSN: 2358-2472

Incisions were closed routinely.

Sample processing and histopathologic evaluation

Liver specimens were measured (length × width) macroscopically with a sterile ruler. The difference between macroscopic and microscopic areas is attributed to tissue shrinkage during formalin fixation and histological processing, a well-documented phenomenon (FANTINATTI et al., 2003). All samples were fixed in 10% neutral buffered formalin for at least 24 hours, processed routinely, embedded in paraffin, sectioned at 5 µm, and stained with hematoxylin and eosin (H&E).

A single board-certified veterinary pathologist, blinded to the sample group, evaluated all slides. The assessment included:

Specimen quality: graded as 0 (Poor, severe artifacts preclude evaluation), 1 (Fair, moderate artifacts but diagnosis possible), 2 (Good, mild artifacts), or 3 (Excellent, minimal to no artifacts).

Diagnostic sufficiency: graded as 0 (Insufficient, inadequate tissue for diagnosis), 1 (Partially sufficient, limited but definitive diagnosis possible), or 2 (Fully sufficient, ample tissue for comprehensive diagnosis).

Edge artifact evaluation: The presence of crushing or compression at the tissue edges was quantified using digital photomicrographs taken at 50x magnification and analyzed with ImageJ software (National Institutes of Health). The total tissue area and the area affected by crushing artifacts were measured in cm^2. The ratio of the crushing area to the whole tissue area was calculated as a percentage.

Portal space count: The number of intact portal spaces was systematically counted in each H&E-stained section to assess architectural adequacy (KEMP et al., 2015).

Statistical analysis

Data were analyzed using the BioStat 5.3 statistical package. Complication rates and macroscopic findings were described by frequency and percentage. Continuous data, such as procedural time and area measurements, were expressed as mean ± standard deviation. The normality of data distribution was assessed with the Shapiro-Wilk test. In the cadaveric phase, comparisons of microscopic area, quality scores, and edge artifact scores between Bf1 and Cf1 groups were performed using a paired t-test and the Wilcoxon signed-rank test, as appropriate. A significance level of $p < 0.05$ was adopted for all analyses.

Revista ARACÊ
ISSN: 2358-2472

3 RESULTS

A ex vivo cadaveric phase

The initial phase of the study aimed to compare the sample quality obtained with Babcock forceps (Bf1) against conventional biopsy forceps (Cf1) in a controlled, *ex vivo* bovine fetus model. This model was selected for its ability to provide tissues of comparable scale to canine patients, allowing for the collection of multiple paired samples for direct comparison, a methodology supported by previous studies evaluating laparoscopic techniques (BUOTE; LOFTUS; MILLER, 2022; DOBBERSTEIN et al., 2024). All collected samples (n=6 per group) exhibited a reddish-brown colour and firm consistency, though a degree of crumbing under pressure was noted, an expected finding attributed to post-mortem autolytic changes.

Both techniques yielded samples of high quality and sufficient size for histopathological analysis (Figure 2 and Figure 3). The area of the fragments and the degree of edge artifacts were comparable, indicating that the Babcock forceps could procure tissue samples of similar gross dimensions to the conventional instrument in this model.

As detailed in Table 1, no statistically significant differences were observed between the two groups for any of the evaluated parameters.

Table 1

Evaluation of laparoscopic liver biopsies performed using conventional biopsy forceps (Cf1) and Babcock forceps (Bf1) in the cadaveric model

Variables	Cf1	Bf1	p-value
Quality	2.8 ± 0.4	2.4 ± 0.5	0.1797
Size for analysis	2 ± 0	2 ± 0	1
edge artefacts	1.4 ± 0.5	1.8 ± 0.4	0.1797
Area (cm^2)	0.40 + 0.2	0.36 + 0.2	0.1466

Caption: Data for quality, size, and edge destruction were subjected to the Wilcoxon Test. Area data were subjected to the t-test.

In vivo clinical phase

Based on the promising ex vivo results, the technique was advanced to clinical evaluation in six canine patients (Bf2). The mean total procedural time from initial incision to closure was 13.2 ± 2.4 minutes. Critically, no intraoperative hemorrhage or other complications were observed at any biopsy site during the laparoscopic observation period

Revista ARACÊ
ISSN: 2358-2472

following sample collection (Figure 1C). The absence of bleeding is attributed to the technique itself, which involves grasping the tissue for five seconds, likely inducing localized ischemia and vascular stasis prior to excision with the sharp trocar sheath, resulting in a clean cut with minimal vascular trauma.

The samples obtained were deemed diagnostically adequate. A notable observation was the difference between the macroscopic area measured fresh (0.19 ± 0.08 cm²) and the microscopic area measured after fixation and processing (0.43 ± 0.02 cm²). This discrepancy is consistent with well-documented tissue shrinkage that occurs during formalin fixation and paraffin embedding (FANTINATTI et al., 2003). The quantitative analysis of crushing artifacts revealed that 21.62% of the total tissue area was affected, a finding that was statistically significant ($p < 0.0001$) when comparing the crushed area to the total viable tissue area. Despite these edge artifacts, the samples contained a mean of 32.62 ± 30.34 intact portal spaces, providing ample architectural context for histopathological evaluation (KEMP et al., 2015).

Table 2

Evaluation of laparoscopic liver biopsies performed using Babcock forceps (Bf2) in canine patients

Variables	Bf2
Procedural time (min.)	13.2 ± 2.4
Macroscopic area (cm^2)	0.19 ± 0.08
Microscopic area (cm^2)	0.43 ± 0.02
edge artefacts area (cm^2)	0.09 ± 0.02
Amount of portal space were	32.62 ± 30.34
Ratio of crushing area to whole tissue	21.62%

The initial and preliminary data of the study, demonstrating the viability of the babcook forceps for collecting liver samples for histopathology, supported the decision to collect liver samples with a babbook clamp from six patients with liver disorders treated at the veterinary hospital of the Federal University of Pará. Thus, liver samples were successfully collected from six canine patients treated in the routine clinical setting. The conclusive diagnoses were achieved through histopathological analysis of the obtained samples, as described below.

The histopathological analysis of the samples revealed distinct tissue patterns

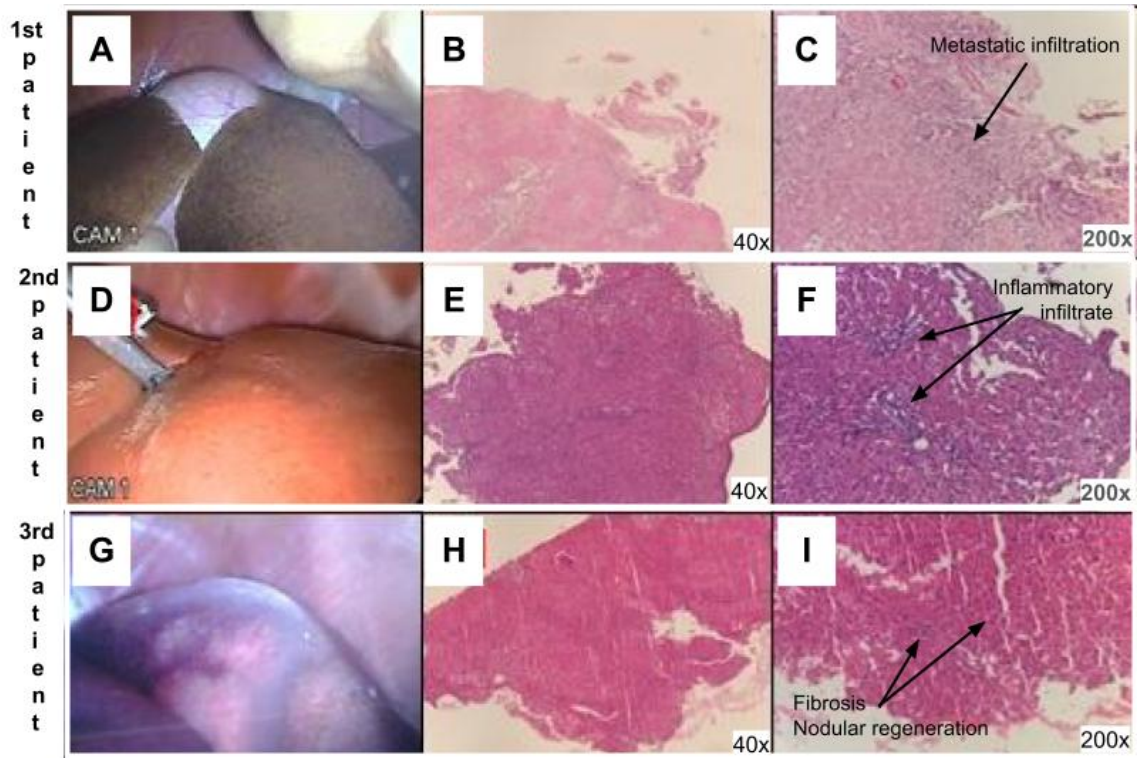
Revista ARACÊ
ISSN: 2358-2472

corresponding to specific diagnoses. In the first patient, the histological sections showed a pattern consistent with metastatic infiltration, leading to the diagnosis of metastatic exocrine pancreatic carcinoma (Figures 2-A, 2-B, 2-C). For the second patient, the microscopic evaluation identified a suppurative inflammatory process within the biliary tract, resulting in the diagnosis of moderate multifocal suppurative cholangitis (Figures 2-D, 2-E, 2-F). The third patient's samples displayed architectural distortion with fibrosis and nodular regeneration, characterizing chronic liver disease associated with moderate portal fibrosis and regenerative nodules (Figures 2-G, 2-H, 2-I).

Revista ARACÊ
ISSN: 2358-2472

Figure 2

Intraoperative laparoscopic views and corresponding histopathological sections of liver samples obtained with Babcock forceps from the first three canine patients. Macroscopic and microscopic images are displayed for each case. First Case (A, B, C): (A) Laparoscopic view of the liver surface. (B) Photomicrograph of the liver sample (H&E stain, 40x magnification) showing a pattern consistent with metastatic infiltration. (C) Higher magnification (200x) of the same sample, confirming the diagnosis of metastatic exocrine pancreatic carcinoma. Second Case (D, E, F): (D) Laparoscopic view of the liver. (E) Photomicrograph (H&E, 40x) revealing a suppurative inflammatory process within the biliary tract. (F) Higher magnification (200x) leading to the diagnosis of moderate multifocal suppurative cholangitis. Third Case (G, H, I): (G) Laparoscopic view of the liver. (H) Photomicrograph (H&E, 40x) displaying architectural distortion with fibrosis and nodular regeneration. (I) Higher magnification (200x) characterizing chronic liver disease associated with moderate portal fibrosis and regenerative nodule



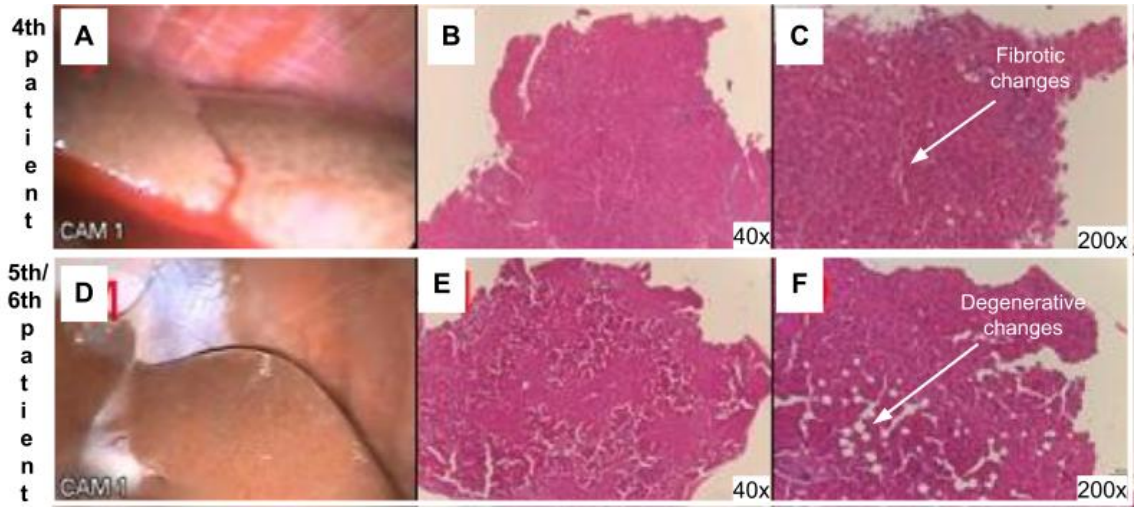
In the fourth patient, the histopathology revealed steatosis, cholestasis, and fibrotic changes, confirming chronic liver disease associated with moderate portal fibrosis, steatosis, and cholestasis (Figures 3-A, 3-B, 3-C). Finally, the fifth and sixth patients presented similar microscopic findings of chronic degenerative changes, leading to the diagnosis of chronic

Revista ARACÊ
ISSN: 2358-2472

degenerative liver disease (Figures 3-D, 3-E, 3-F).

Figure 3

Intraoperative laparoscopic views and corresponding histopathological sections of liver samples obtained with Babcock forceps from the fourth, fifth, and sixth canine patients. Fourth Case (A, B, C): (A) Laparoscopic view of the liver. (B) Photomicrograph of the liver sample (H&E stain, 40x magnification) showing steatosis and fibrotic changes. (C) Higher magnification (200x) confirming chronic liver disease associated with moderate portal fibrosis, steatosis, and cholestasis. Fifth and Sixth Cases (D, E, F): (D) Laparoscopic view of the liver (representative of both patients). (E) Photomicrograph (H&E, 40x) demonstrating chronic degenerative changes. (F) Higher magnification (200x) leading to the diagnosis of chronic degenerative liver disease for both patients



Postoperatively, the first patient died five days after the procedure, possibly due to an advanced degree of pre-existing hepatic impairment. The third patient developed a small seroma at the second trocar incision site two days post-procedure, which was resolved with drainage and a compressive bandage.

4 DISCUSSIONS

O This study demonstrates that laparoscopic liver biopsy using Babcock forceps is a feasible, efficient, and safe technique for obtaining diagnostic tissue samples in dogs. The two-phase study design, beginning with an ex vivo cadaveric model, provided a crucial platform for standardizing the technique and conducting an initial comparative assessment of sample quality before proceeding to clinical patients, a methodological approach

Revista ARACÊ
ISSN: 2358-2472

supported by recent literature (BUOTE; LOFTUS; MILLER, 2022; DOBBERSTEIN et al., 2024). While the cadaveric model has limitations, primarily the inability to assess hemorrhage due to the absence of blood pressure and coagulation, it remains a validated tool for evaluating the procedural steps and the gross and histological characteristics of the obtained samples (SINGH; CASE, 2022). The lack of significant difference in sample quality and area between Babcock and conventional biopsy forceps in the cadaveric phase provided the necessary justification to advance the technique to clinical testing.

In the clinical phase, the most significant finding was the complete absence of intraoperative hemorrhage. This contrasts with Reviewer 2's concern that Babcock forceps may induce bleeding. We posit that the technique itself mitigates this risk. The act of grasping the tissue for five seconds prior to transection likely causes localized vascular compression and stasis, and the subsequent clean incision with the sharp trocar sheath minimizes tissue tearing. This mechanism appears to be effective, as no bleeding was observed despite the procurement of diagnostically sufficient tissue. The mean procedural time of 13.2 minutes was notably shorter than the average of 26 minutes reported in other studies (FERNANDEZ et al., 2017; PETRE et al., 2012). This efficiency can be attributed to the technique's simplicity, utilizing a common instrument (Babcock forceps) in a straightforward two-step process (grasp and cut), potentially eliminating the need for instrument changes or more complex maneuvers like suture ligation.

A key aspect of sample quality is the preservation of tissue architecture for histopathological interpretation. The crushing artifact observed at the edges of the samples (21.62% of the total area) is an expected consequence of using a grasping instrument. However, this level of artifact was significantly lower than the 70% crushing reported with hot-biopsy forceps, which can render some samples non-diagnostic (SOUZA et al., 2012). It was also comparable to the 14.6% crushing caused by electrocautery forceps (FANTINATTI et al., 2003). Importantly, in our study, the crushing was confined to the tissue edges and did not compromise the diagnostic quality of the central parenchyma, as evidenced by the ability to reach a definitive morphological diagnosis in all cases. While techniques like pre-tied ligatures (endoloop) can yield larger samples with minimal crushing (FERNANDEZ et al., 2017), they may be more technically demanding or require specialized, costly equipment.

The diagnostic adequacy of the Babcock technique is underscored by the number of portal spaces obtained (32.62 ± 30.34) and the range of conditions successfully diagnosed, from neoplastic (metastatic carcinoma) to inflammatory (cholangitis) and chronic

Revista ARACÊ
ISSN: 2358-2472

degenerative diseases. Although an optimal number of portal spaces for canine liver biopsies has not been standardized, our samples provided ample architectural context, aligning with the principle that sample quality is paramount (KEMP et al., 2015). The combination of direct laparoscopic visualization of the liver surface with the histopathological findings from viable biopsy fragments creates a powerful diagnostic synergy, allowing for targeted sampling and comprehensive assessment (MCDEVITT et al., 2016).

This study has limitations, including the small clinical sample size and the single-center design. The use of a bovine fetus cadaveric model, while practical for initial testing, does not perfectly replicate the biomechanical properties of a pathologic canine liver in vivo. Furthermore, the subjective component of the quality grading scale, despite the blinding of the pathologist, is a recognized limitation. Future studies with larger case numbers and direct comparison to other established techniques in a randomized clinical trial are warranted to solidify these findings.

Notwithstanding these limitations, the findings hold clinical significance. The Babcock forceps technique offers a practical and accessible alternative for veterinary surgeons. In settings where specialized biopsy forceps are unavailable, this technique enables the performance of a high-quality laparoscopic liver biopsy using a standard, multi-purpose instrument found in most laparoscopic sets. This can potentially increase access to minimally invasive diagnostic procedures for a greater number of patients.

5 CONCLUSIONS

A This study demonstrates that the laparoscopic liver biopsy technique using Babcock forceps is a feasible, safe, and efficient method for obtaining diagnostic tissue samples in dogs. Contrary to the concern that Babcock forceps may induce hemorrhage, our in vivo findings confirmed an absence of intraoperative bleeding, which we attribute to the technique of grasping the tissue to induce localized ischemia before a clean transection with the trocar sheath. The two-phase study design, beginning with a bovine fetus cadaveric model for initial technique comparison and standardization (BUOTE; LOFTUS; MILLER, 2022), proved to be a valid approach for developing a novel clinical technique before its application in patients.

The technique yielded samples of consistent quality, with a crushing artifact profile (21.62%) that, while present, was manageable and did not preclude a definitive histopathological diagnosis for a range of liver diseases, including neoplasia, cholangitis, and

Revista ARACÊ
ISSN: 2358-2472

chronic hepatopathies. The procedural time was short, and the method utilized a common laparoscopic instrument, underscoring its practicality and accessibility.

Therefore, this investigation holds clear clinical significance. The Babcock forceps technique provides a viable and cost-effective alternative for veterinary surgeons, particularly in settings where specialized biopsy forceps are not available. It enables the performance of high-quality, minimally invasive liver biopsies, thereby expanding diagnostic capabilities. Future studies with a larger sample size are recommended to further validate these findings and directly compare this technique against other established methods.

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