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ESTRATÉGIAS PARA MELHORIA NA EFICIÊNCIA E COMUNICAÇÃO DOS LABORATÓRIOS DE DIAGNÓSTICO VETERINÁRIO

GISELE CALDAS BONATO

Campo Grande – MS 2024 **GISELE CALDAS BONATO**

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Strategies for improving the efficiency and communication of veterinary diagnosis laboratories

GISELE CALDAS BONATO

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Tese apresentada ao Programa de Pós-Graduação em Ciências Veterinárias da Universidade Federal de Mato Grosso do Sul, como requisito parcial para a obtenção do título de Doutora em Ciências Veterinárias.

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DEDICATÓRIA

Primeiramente à Deus, o criador que me deu o maior presente que é a vida. Deus é o centro da minha vida, em que me apoio em tudo que faço, Ele é a minha base. Aos meus pais que me dão todo o apoio, sou imensamente grata por tudo que sempre fizeram por mim, muitas vezes tiraram de si pra me dar. Esse trabalho é fruto de todo o investimento que fizeram em mim, especialmente na minha educação, portanto também é de vocês. A minha família que me dão todo o apoio em todas as áreas da minha vida, sou muito abençoada por ter a minha família sempre presente, minha irmã, minha sobrinha, as tias, tios, primas e primos, meu cunhado e meus avós Antônio e Lígia (in memorian), eu vos amo de todo o meu coração, cada um de vocês é peça fundamental na minha vida. Ao Dr. José Cardoso (in memorian) que foi um pai na veterinária pra mim, tudo que aprendi com você levarei pra sempre comigo, desejo que esteja descansando em um bom lugar junto de Deus. Aos amigos que fiz em Campo Grande, entre eles as meninas do laboratório (Acauane, Helena, Larissa e Thaísa) e o professor Danilo, todos vocês são muito especiais pra mim, me ajudaram a fazer com que dois anos morando longe da família fosse um pouco mais leve e prazeroso, tenho saudades do dia-adia no laboratório com vocês. Além da amizade, aprendi muito mais com vocês sobre patologia, cada dia no laboratório era um aprendizado. A toda equipe do Laboratório de Anatomia Patológica, Raiane, Nicole e Gerson, externo a minha gratidão. Aos professores da pós-graduação que contribuiram ativamente no meu doutorado. Ao professor Ricardo, obrigada por ter me aceitado como sua orientada, por ter me ensinado ainda mais sobre o mundo da patologia, foi realmente muito edificante pra mim a vivência no LAP, espero que eu consiga levar à frente tudo que eu aprendi com vocês, e que eu consiga causar o mesmo impacto na vida profissional das pessoas, assim como vocês fizeram na minha. Agradeço também ao Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) pelo apoio financeiro do meu doutorado.

"Nem olhos viram, nem ouvidos ouviram, nem jamais penetrou o coração humano, o que Deus tem preparado para aqueles que O amam." Bíblia Sagrada 1 Coríntios 2.9

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Fig.10. Edema in the dewlap region in cattle. High quality. The condition occurs as outbreaks in several rural properties and affects many cattle, usually after an intense cold. Recovery is spontaneous, but this condition must be distinguished from hepatogenous photosensitization caused by intoxication by *Brachiaria* spp., *Enterolobium contortisiliquum*, and *Stryphnodendron fissuratum*, in addition to heart failure and its causes. The initial suspicion of intoxication by *Brachiaria* sp. was ruled out based on clinical course and condition, with spontaneous recovery and without photodermatitis. Serum levels of the enzyme gamma-glutamyl transferase (GGT) and aspartate aminotransferase (AST) were within physiological values and were considered complementary diagnostic criteria. Image provided by the veterinarian Guilherme Dutra, Rodrigo Spengler and Luiz Carlos Louzada Ferreira.

CAPÍTULO 3

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CAPÍTULO 4

The use of WhatsApp to identify previously undiagnosed and underreported ruminant and equine diseases in Mato Grosso do Sul

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CAPÍTULO 4

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RESUMO

BONATO, G.C. ESTRATÉGIAS PARA MELHORIA NA EFICIÊNCIA E COMUNICAÇÃO DOS LABORATÓRIOS DE DIAGNÓSTICO VETERINÁRIO. 2024. Tese de Doutorado - Programa de Pós-Graduação em Ciências Veterinárias. Faculdade de Medicina Veterinária e Zootecnia, Universidade Federal de Mato Grosso do Sul, Campo Grande, MS, 2024.

Os Laboratórios de Diagnóstico Veterinário (LDVs) desempenham um papel crucial para pesquisa, desenvolvimento e inovação para o controle de doenças. Reduzir as perdas causadas por problemas sanitários é fundamental para alcançar a eficiência econômica e dos serviços dos laboratórios e veterinário. Os dados gerados a partir do serviço dos laboratórios contribuem com veterinários de campo e instituições de ensino por meio do intercâmbio dessas informações. A realização de estudos retrospectivos ajudam a identificar as causas e as condições em que ocorrem as doenças, porém há desafios como a coleta de amostras inadequadas e a ausência de informações. O objetivo desse estudo foi implementar estratégias para melhorar a eficiência e comunicação dos laboratórios de diagnóstico veterinário. O aplicativo de mensagens Whatsapp® foi implementado para melhorar a comunicação entre LDVs, veterinários e produtores. Isso incluiu o envio de imagens de casos clínicos, troca de informações em um grupo específico e publicações técnicas para difusão do conhecimento. A partir da estratégia implementada foram gerados artigos científicos, capítulos de livros e notas técnicas. Isso fortalece o conhecimento técnico e promove a educação continuada dos profissionais, além de aumentar a colaboração com o setor. A utilização de ferramentas modernas de comunicação, como o Whatsapp, demonstrou ser crucial para aprimorar a eficiência dos LDVs. A metodologia implementada foi eficiente em aumentar o número de diagnósticos conclusivos em relação ao ano anterior e em identificar doenças subnotificadas, monitorar surtos e identificar práticas veterinárias equivocadas pelos profissionais e corrigi-las. Os materiais obtidos são uma importante fonte para o laboratório que podem ser transformados em publicações científicas e técnicas.

Palavras-chave: diagnóstico veterinário; mídias sociais; sanidade animal.

ABSTRACT

BONATO, G.C. ESTRATÉGIAS PARA MELHORIA NA EFICIÊNCIA E COMUNICAÇÃO DOS LABORATÓRIOS DE DIAGNÓSTICO VETERINÁRIO. 2024. Tese de Doutorado - Programa de Pós-Graduação em Ciências Veterinárias. Faculdade de Medicina Veterinária e Zootecnia, Universidade Federal de Mato Grosso do Sul, Campo Grande, MS, 2024.

Veterinary Diagnostic Laboratories (VDLs) play a crucial role in research, development and innovation for disease control. Reducing losses caused by health problems is fundamental to achieving economic efficiency and the efficiency of laboratory and veterinary services. The data generated from laboratory services contributes to field veterinarians and educational institutions through the exchange of this information. Conducting retrospective studies helps to identify the causes and conditions in which diseases occur, but there are challenges such as inadequate sample collection and lack of information. The aim of this study was to implement strategies to improve the efficiency and communication of veterinary diagnostic laboratories. The Whatsapp® messaging app was implemented to improve communication between LDVs, veterinarians and producers. This included sending images of clinical cases, exchanging information in a specific group and technical publications to disseminate knowledge. From the strategy implemented, scientific articles, book chapters and technical notes were generated. This strengthens technical knowledge and promotes continuing education for professionals, as well as increasing collaboration with the sector. The use of modern communication tools, such as WhatsApp, has proved crucial in improving the efficiency of LDVs. The methodology implemented was effective in increasing the number of conclusive diagnoses compared to the previous year and in identifying under-reported diseases, monitoring outbreaks and identifying wrong veterinary practices by professionals and correcting them. The materials obtained are an important source for the laboratory and can be transformed into scientific and technical publications.

Keywords: veterinary diagnosis; social media; animal health.

CAPÍTULO 1

1. INTRODUÇÃO

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Com o aumento da demanda, da competitividade internacional e da 5 exigência do mercado consumidor, a produção de carne bovina passou a seguir 6 padrões mais rígidos nos últimos anos. O aumento da produtividade só foi possível 7 8 com as pesquisas desenvolvidas em universidades, iniciativa privada, instituições 9 de pesquisa e Laboratórios de Diagnóstico Veterinário (LDVs) (SCHULZ et al 2018). Para buscar a excelência técnica e econômica torna-se obrigatório fortalecer a 10 11 pesquisa, desenvolvimento e inovação para alavancar a pecuária de corte no cenário mundial. É mandatório reduzir as perdas causadas por problemas sanitários 12 13 evitáveis, o que só é possível com o aumento da eficiência do diagnóstico e o estabelecimento de medidas eficientes de prevenção e controle das doenças 14 15 (LEMOS et al 2022). O atendimento a essas exigências requer a incorporação de 16 novas práticas e de novas tecnologias no processo de diagnóstico e tomada de 17 decisão para o controle de doenças (CARTER e SMITH 2021, EL-SAYED e KAMEL 2018). 18

Nesse sentido, os laboratórios de diagnóstico veterinário (LDVs) cumprem um 19 papel fundamental, pois dedicam-se ao estudo das doenças dos animais desde a 20 ótica da epidemiologia, das causas determinantes das mesmas e do seu controle 21 22 (SALIKI 2000). A atuação dos LDVs baseada na coleta e análise de dados de forma padronizada e sistematizada permite a elaboração de material de divulgação 23 destinado, principalmente, aos médicos veterinários de campo, pois os dados 24 oriundos dos laboratórios fornecem importante orientação sobre as principais 25 26 enfermidades que ocorrem na sua área de atuação (LUCENA et al. 2010, RISSI et al. 2010). O intercâmbio de informações entre os órgãos oficiais de defesa sanitária 27 e veterinários da iniciativa privada colabora para a identificação e controle de 28 doenças. Essas informações podem ser repassadas a instituições de ensino e 29 30 pesquisa, como as universidades, e assim contribuir na formação de novos profissionais e no processo de educação continuada. 31

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Nesse contexto, os LDVs elaboram estudos retrospectivos a fim de estudar

uma determinada população em um período de tempo, assim, auxiliam na redução 33 34 dos prejuízos causados pelas doenças, pois o controle eficiente das mesmas 35 depende da identificação correta da causa e também das condições de ocorrência das doenças (ANTONIASSI et al 2013, CARMO et al 2011, HECKLER et al 2018, 36 HELMAN 2020, LUCENA et al. 2010, MOLOSSI et al. 2021, PUPIN et al. 2019, 37 38 RISSI et al 2010, RONDELLI et al. 2017, SPRENGER et al. 2015). No entanto, vários fatores limitantes levam a um elevado número de diagnósticos inconclusivos 39 e que reduzem a eficiência desses laboratórios. Como principais causas são 40 descritas o envio inadequado de amostras, seja por mau acondicionamento ou não 41 42 envio de partes anatômicas pertinentes ao diagnóstico e a falta de informações clínicas e epidemiológicas (BASSUINO et al 2016, LUCENA et al 2010, PUPIN et 43 44 al 2019, RONDELLI et al. 2017). Em parte, pode se considerar que estes pontos 45 resultam da forma de comunicação dos LDVs, muitas vezes realizada por meio de publicações em periódicos científicos, aos quais seu público alvo nem sempre tem 46 47 acesso. Nesse sentido, iniciativas adotadas como a publicação de material 48 bibliográfico impresso, baseado em situações reais da rotina do LAP/FAMEZ e a 49 criação de um canal de comunicação direta entre o LAP/FAMEZ e os médicos 50 veterinários e produtores rurais, tiveram resultados promissores (LEMOS et al 2022, 51 WOODARD et al 2021).

Porém, essas iniciativas foram adotadas em caráter experimental e destinadas a um público restrito. Sua massificação representará um importante avanço na forma de atuação do LAP/FAMEZ. Além de aumentar a eficiência do diagnóstico, a comunicação em tempo real, possibilita a tomada de decisão em curto espaço de tempo, o que em situações de campo é fundamental para redução de perdas.

Ressalta-se que existe uma lacuna na forma de detecção e controle de doenças, especialmente aquelas que podem não ser notificadas devido a diversas razões, como doenças com tratamentos eficazes, mortes e sintomas semelhantes a outras doenças. Um dos pontos cruciais é a necessidade de critérios bem definidos para o diagnóstico correto, evitando assim a adoção de protocolos de tratamento custosos e desnecessários. Isso não apenas economiza recursos, mas

também melhora a eficiência do controle de doencas. Além disso, o 64 acompanhamento pós-diagnóstico das propriedades é enfatizado como uma prática 65 66 essencial. Isso envolve avaliar a eficácia das medidas recomendadas após o diagnóstico inicial, garantindo que as intervenções adotadas sejam realmente 67 eficazes na redução da prevalência e na mitigação dos impactos das doenças. 68 69 Essas medidas não se aplicam apenas às doenças já sob programas oficiais de controle, mas também a outras doenças que podem ser confundidas com aquelas 70 monitoradas de forma oficial, servindo como alertas importantes para a ocorrência 71 72 dessas doenças.

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74 **2. OBJETIVO**

75 2.1. Objetivo geral

Implementar estratégias para melhoria da eficiência e da comunicação em
 laboratórios de diagnóstico veterinário.

78 2.2. Objetivos específicos

- 1. Uso da tecnologia de Informação (Whatsapp®) como ferramenta para melhoria
 da comunicação e aumento da eficiência do atendimento em casos de
 ocorrências sanitárias em animais de produção;
- 2. otimizar a transferência de conhecimento científico e tecnológico produzido pelo
 LAP/FAMEZ para seu público alvo.

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85 **3. METODOLOGIA**

86 No ano de 2022, foi implementado um grupo de Whatsapp®, no qual foram incluídos médicos veterinários dos serviços público e privado. Inicialmente foram 87 88 adicionados somente os veterinários que já tinham feito contato por Whatsapp® ou 89 utilizado o serviço do laboratório em algum momento, sendo 34 participantes, e, conforme outros veterinários utilizavam o serviço do laboratório eram incluídos 90 nesse grupo. Até o mês de julho de 2024, o grupo contava com 320 participantes. 91 92 Neste grupo, os participantes tinham acesso aos casos e às dúvidas levantadas 93 pelos profissionais.

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Todos os participantes tiveram acesso a um número de telefone celular

disponibilizado pelo Laboratório de Anatomia Patológica da UFMS (LAP/UFMS) e,
por esse número, eles poderiam encaminhar imagens e informações sobre casos
clínicos que estavam acompanhando no momento, dúvidas sobre doenças de
animais de produção, bem como solicitar orientações sobre coleta e envio de
amostras de órgãos para os laboratórios de diagnóstico ou agendamentos de visitas
da equipe do laboratório à propriedade em questão.

A partir do ano de 2022, houve reuniões presenciais anuais para divulgação dos resultados dos diagnósticos realizados pela equipe do LAP/UFMS durante aquele ano, os participantes do grupo tinham prioridade para participação nesses eventos. Também foram elaborados boletins técnicos a partir desses resultados. Os boletins estão disponíveis em https://ppgcivet.ufms.br/boletim-de-diagnostico-dolaboratorio-de-anatomia-patologica/

Foram elaboradas notas técnicas sobre as principais causas de morte de animais de produção diagnosticadas pela equipe do LAP/UFMS, as quais eram periodicamente postadas no grupo. As nota técnicas estão disponíveis em https://ppgcivet.ufms.br/notas-tecnicas-em-sanidade-de-animais-de-producao/

111 O grupo também podia ser utilizado para prospecção de informações sobre 112 doenças importantes na região. Os materiais desenvolvidos foram incorporados à 113 rotina do laboratório e estão disponíveis para consulta no LAP-UFMS e no site da 114 pós-graduação da Faculdade de Medicina Veterinária e Zootecnia-UFMS.

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116 **4. RESULTADOS**

Os dados de todos os contatos provenientes do Whatsapp® foram 117 armazenados junto à uma base de dados do LAP-UFMS e posteriormente 118 119 analisados. Os contatos foram divididos em grupos em: a) contatos que resultaram 120 no envio de material para exames laboratoriais e b) contatos que não houve remessa de material. Todos os contatos com remessa de material foram registrados em fichas 121 do LAP-UFMS. Os dados obtidos originaram a publicação de três artigos, duas notas 122 técnicas e um capítulo do boletim anual do LAP-UFMS, os quais estão descritos nos 123 124 capítulos de 2 a 5.

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201 CAPÍTULO 2 – Use of smartphone messaging applications to 202 increase diagnostic efficiency in veterinary diagnostic laboratories

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Use of smartphone messaging applications to increase diagnostic efficiency in veterinary diagnostic laboratories¹

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ABSTRACT.- Lemos R.A.A., Guizelini C.C., Silva T.X., Souza L.L., Fonseca H.C.F., Bonato G.C., Gomes D.C. & Pupin R.C. 2023. **Use of smartphone messaging applications to increase diagnostic efficiency in veterinary diagnostic laboratories**. *Pesquisa Veterinária Brasileira 43:e07226, 2023*. Laboratório de Anatomia Patológica, Faculdade de Medicina Veterinária e Zootecnia, Universidade Federal de Mato Grosso do Sul, Av. Sen. Filinto Müler 2443, Campo Grande, MS 79070-900, Brazil. E-mail: <u>ricardo.lemos@ufms.br</u>

Many inconclusive diagnoses have been reported in studies carried out in veterinary diagnostic laboratories (VDLs). These inconclusive diagnoses are often associated with field veterinarians sending tissue samples and carcasses for histopathological examination. The use of social media in care has the potential as a tool for improvements in animal health. We evaluated the improvement in the rate of conclusive diagnoses in the VDL when using a messaging application for smartphones as a guidance tool for field veterinarians to refer animals and sample tissues for necropsy and histopathology. Veterinarians contacted the VDL team via a messaging app and forwarded videos, images, and clinical-epidemiological history. Based on this information, guidelines were created in real-time for contacting professionals about methods for collecting samples during necropsy and making diagnoses and scheduling visits to properties. The data obtained showed that smartphone messaging applications could be useful tools to expand the performance of VDLs and improve their diagnostic efficiency, especially when sending samples of animals that died is impossible due to certain sanitary conditions.

INDEX TERMS: Smartphone messaging applications, diagnostic laboratories, animal production, veterinary pathology.

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RESUMO.- [Uso de aplicativos de mensagens de smartphones para

aumentar a eficiência diagnóstica em laboratórios de diagnóstico veterinário.] Muitos diagnósticos inconclusivos têm sido relatados em estudos realizados em laboratórios de diagnóstico veterinário (LDVs). Esses diagnósticos inconclusivos são frequentemente associados a veterinários de campo que enviam amostras de tecidos e carcaças para exame histopatológico. O uso das mídias sociais com cuidado tem potencial como ferramenta para melhorias na saúde animal. Avaliamos a melhora na taxa de diagnósticos conclusivos no LDV ao utilizar um aplicativo de mensagens para smartphones como ferramenta de orientação para veterinários de campo encaminharem animais e amostras de tecidos para necropsia e histopatologia. Os veterinários entraram em contato com a equipe do L por meio de um aplicativo de mensagens e encaminharam vídeos, imagens e histórico clínico-epidemiológico. Com base nessas informações, foram criadas orientações em tempo real para contato dos profissionais sobre métodos de coleta de amostras durante a necropsia e realização de diagnósticos, além do agendamento de visitas às propriedades. Os dados obtidos mostraram que os aplicativos de mensagens para smartphones podem ser ferramentas úteis para ampliar o desempenho dos VDLs e melhorar sua eficiência diagnóstica, principalmente quando o envio de amostras de animais que morreram é impossível devido a determinadas condições sanitárias.

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301 302 TERMOS DE INDEXAÇÃO: Aplicativos de mensagem, laboratórios de diagnóstico, produção animal, patologia veterinária.

INTRODUCTION

Veterinary diagnostic laboratories (VDLs) are a fundamental part of the integrated diagnostic
system in animal health, as they work in the diagnosis, prevention, control, and treatment of diseases,
in addition to contributing to the development and improvement of diagnostic techniques. This set of
actions results in an important economic impact on livestock activity by reducing losses caused by
diseases (Schulz et al. 2018).

Retrospective studies carried out by these VDLs in Brazil (Lucena et al. 2010, Sprenger et al. 2015, Mello et al. 2017, Rondelli et al. 2017, Pupin et al. 2019, Molossi et al. 2021) indicate that 72.9% to 89.94% of inconclusive diagnoses are obtained from samples sent by field veterinarians. The main reasons for this are the sending of tissues in insufficient quantity or an advanced stage of autolysis, the non-referral of organs possibly associated with the reported clinical signs, and the lack of epidemiological and clinicopathological information.

287 Therefore, to reduce the number of inconclusive diagnoses resulting from failures in forwarding 288 materials to the VDLs, it is necessary to implement measures to minimize these problems. The use of 289 social media for teaching and improving the speed of diagnoses in Veterinary Medicine (Bernardo et al. 290 2013, Tenhaven et al. 2013, Mekaru & Brownstein 2014, Haustein et al. 2015, Kedrowicz et al. 2016, 291 Englar 2017, Ober 2019, Saadeh et al. 2021, Sellers et al. 2021, Trittmacher et al. 2021, Woodard et al. 292 2021) and telemedicine (Farahani & Pantanowitz 2015, Bertram & Klopfleisch 2017) grows 293 exponentially. The use of smartphones and media during visits to rural properties and in the guidance 294 to veterinarians on the procedures necessary for sending samples to the VDLs, respectively, showed the 295 potential to stimulate improvements in animal health (Karimuribo et al. 2016, Miltenburg et al. 2021). 296 However, we could find no specific reports addressing the role of messaging applications designed for 297 smartphones as a tool to aid field veterinarians in the elaboration of diagnoses.

This study aimed to evaluate the efficiency of smartphone messaging applications as a tool to guide and direct field veterinarians during the collection and sending of samples to assess whether there is an improvement in the rate of conclusive diagnoses in VDLs.

MATERIALS AND METHODS

For data collection, the team from the "Laboratório de Anatomia Patológica" of the "Universidade
 Federal de Mato Grosso do Sul" (LAP- UFMS) provided veterinarians with a cell phone number
 registered in a messaging application, asking them to share this number with other veterinarians. From
 January 2020 to November 2021, all clients who referred clinical cases via text or audio messages,

images, and videos on this digital platform were considered in the study.

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The messages received were cataloged and evaluated in terms of the municipality where the clinical cases occurred, the quality of the images and videos referring to the clinical cases and lesions found during the necropsy of the animals, if there was a referral at LAP- UFMS of carcasses or tissues samples from animals necropsied, and their respective number of conclusive and inconclusive diagnoses.

313 For the evaluation of image quality, only those sent via the messaging application by field 314 veterinarians were considered. The images were classified into three groups: a) poor quality: there was 315 no concrete visualization of clinical signs and/or macroscopic lesions from the images; b) good quality: 316 it was possible, with some graphic difficulty, to determine the clinical signs and/or the macroscopic 317 lesions present in the images and classify them in a diagnostic category (e.g., neurological syndromes, 318 myopathies, and hepatotoxic or cardiovascular diseases); and c) high quality: when the images made it 319 possible to accurately identify the clinical signs and macroscopic lesions (e.g., jaundice, pneumonia, and 320 myonecrosis).

The conclusive diagnoses were based on identifying the etiologic agent in tissue and fluid samples from the necropsied animals, either by histopathological examination or other complementary exams. When the images associated with the epidemiological and clinical history were sufficient to determine the cause of death of the animal, the diagnosis was also conclusive. When macroscopic lesions were sufficient to cause clinical signs and/or death, but the etiological agent was not determined, the morphologic diagnosis was instituted as the final diagnosis. Diagnoses were considered inconclusive when it was not possible to identify lesions that could be associated with the clinicopathological aspects.

RESULTS

During the period evaluated, 411 samples of cattle, 80 horses, 16 sheep, 11 pigs, and two goats were sent to the LAP-UFMS; of this total, 88 (21.4%) were included in this study. The farms of the assisted animals are located in the states of Mato Grosso do Sul (31 municipalities), Mato Grosso, Goiás, Bahia and Pará (Table 1) and have minimum and maximum distances to the physical headquarters of the LAP-UFMS of 58.5km and 2,890km, respectively.

A total of 95 veterinarians contacted the cell phone number provided; of these, 35 (36.84%) sent carcasses to the LAP-UFMS for a necropsy by the specialized team, and 25 (26.31%) performed the necropsy directly in the field, sending organ samples for histopathological exam and other complementary tests, such as microbiological culture. The remaining 35 (36.84%) did not send tissue samples or carcasses but recorded images of clinical signs and provided some epidemiological information about the case. Among the veterinarians who forwarded tissue samples from animals or cadavers to the LAP-UFMS (60), 34 used this service for the first time and made new shipments later.

The LAP-UFMS team performed 45 necropsies, 32 (71.11%) of which had a conclusive diagnosis, two (4.44%) had a suggestive diagnosis, and 11 (24.44%) had inconclusive diagnoses. In addition to frequently diagnosed conditions (Table 2), this study included – with a frequency of 3.12% (1/32) of each disease – nutritional myopathy, poisoning by Enterolobium contortisiliquum, urea and ionophore antibiotics, bacterial discospondylitis, malnutrition, hypovolemic shock secondary to severe anemia due to high tick infestation, traumatic reticulopericarditis, and co-infection between Anaplasma sp. and Salmonella sp.

349There were 39 samples collected in necropsies performed by field veterinarians, of which 81.4%350(31), 16.3% (7), and 2.3% (1) had a conclusive diagnosis (Table 1), inconclusive and suggestive of some351condition, respectively. Regarding the 63 contacts who did not send tissue samples or cadavers but sent352images and videos, the inconclusive diagnosis predominated and totaled 68.25% (43/63), mainly due353to the non-performance of necropsies or the not sending samples for histopathology.

The conclusive diagnoses in this category were associated with diseases whose macroscopic lesions and clinical history were sufficient, such as urethral obstruction and intestinal torsion. One of these situations resulted in a visit by the LAP-UFMS team to the property to collect fecal samples for bacterial culture; in 14 other cases (22.22%), the macroscopic images or clinical-epidemiological information allowed the narrowing of the differential diagnoses, such as the suspicion of polioencephalomalacia after treatment with dexamethasone and vitamin B1. Based on the information presented by the veterinarians, other possible diagnoses were poisoning by Brachiaria spp. and Brachiaria radicans (Figs.1-4) and Stryphnodendron fissuratum (Figs.5-7), hypothermia, sodium deficiency, and ruminal acidosis (Figs.8-9). Sporadically diagnosed diseases included suppurative discospondylitis, bovine herpesvirus (BoHV) encephalitis, ionophore antibiotic poisoning, and dermatophilosis.

Regarding the carcasses sent for a necropsy to the LAP- UFMS, the inconclusive diagnoses were obtained from severe autolysis (2/10), absence of macroscopic and histological lesions (3/10), and diseases of the reproductive tract (abortions and malformations), in which the etiologic agent could not be identified or there were no macroscopic and microscopic changes (5/10). The organ and tissue samples sent by field veterinarians to the LAP-UFMS that were inconclusive were autolyzed (1/7), suggestive of botulism, but it was not possible to detect botulinum toxin (1/7); there were no macroscopic and microscopic lesions (3/7) or resulted from incomplete sending of samples necessary for diagnoses, such as the absence of brain and spinal cord in situations where neurological diseases were suspected (2/7).

Clinical-pathological suspicions of field veterinarians who referred cadavers or samples of organs
from necropsied animals corresponded to the conclusive diagnosis in approximately 26.7% and 40%
of the cases, respectively. When only images and epidemiological information were sent, the contact's
suspicion was correct in 9.8% of the cases.

Regarding the quality of images and videos sent by contacting veterinarians, most were classified
as good quality (46.42%), followed by poor (44.64%), and high (8.92%). There were situations in which,
even with poor quality, the images were decisive for the diagnosis.

.2	Table 1. Municipalities and states assisted by the LAP-UFMS and their total cases included in this study			
	Municipality/State	in total cuses included in this study	Total (%)	
	Mato Grosso do Sul	Dois Irmãos do Buriti	14 (9.65%)	
		Terenos	14 (9.65%)	
		Campo Grande	13 (9%)	
		Água Clara	12 (8.3%)	
		Paraíso	8 (5.51%)	
		Aquidauana	7 (4.8%)	
		Corumbá	6 (4.13%)	
		Jaraguari	6 (4.13%)	
		Nioaque	6 (4.13%)	
		Coxim	5 (3.44%)	
		Miranda	5 (3.44%)	
		Naviraí	5 (3.44%)	
		Paranaíba	4 (2.75%)	
		Dourados	3 (2.06%)	
		Eldorado	3 (2.06%)	
		Jardim	3 (2.06%)	
		Maracaju	3 (2.06%)	
		Rio Brilhante	3 (2.06%)	
		Rio Negro	3 (2.06%)	
		Bataguassu	2 (1.4%)	
		Camapuã	2 (1.4%)	
		Bandeirantes	1 (0.7%)	
		Bodoquena	1 (0.7%)	
		Caracol	1 (0.7%)	
		Cassilândia	1 (0.7%)	
		Chapadão do Sul	1 (0.7%)	
		Costa Rica	1 (0.7%)	
		Nova Andradina	1 (0.7%)	
		Rochedo	1 (0.7%)	
		Santa Rita do Pardo	1 (0.7%)	
		São Gabriel D'Oeste	1 (0.7%)	
	Pará		4 (2.75%)	
	Bahia		2 (1.4%)	
	Goiás		1 (0.7%)	
	Mato Grosso		1 (0.7%)	

Table 2. Main conclusive diagnoses in farm animals diagnosed by the LAP-UFMS through necropsy performed by the LAP team or by field veterinarians in 2020 and 2021

Necropsy performed by LAP- UFMS team		Necropsy performed by fiel	Necropsy performed by field veterinarian		
Diagnosis	Frequency	Diagnosis	Frequency		
Rabies	1.87% (7/32)	Rabies	42.85% (15/35)		
Botulism	9.37% (3/32)	Bovine respiratory disease	5.71% (2/35)		
Bovine respiratory disease	6.25% (2/32)	Polioencephalomalacia of unknown cause	5.71% (2/35)		
Escherichia coli and Cryptosporidium sp. enteritis	6.25% (2/32)				
Brachiaria sp. poisoning	6.25% (2/32)				
Polioencephalomalacia Bovine herpesvirus encephalitis	6.25% (2/32) 6.25% (2/32)				

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DISCUSSION

The data obtained demonstrate that smartphone messaging applications can be useful tools to
expand the performance of VDLs and improve their diagnostic efficiency (Karimuribo et al. 2016,
Miltenburg et al. 2021), especially when sending samples of animals that died due to a certain condition
is not possible.

422 Approximately 21.5% of the diagnoses in production animals performed by the LAP-UFMS team resulted from contacting veterinarians via messaging application. The replacement of telephone 423 424 contact with electronic means, such as e-mail, creates a certain distance between the user and the VDL. 425 The tool used in this study provided direct and personal communication between the VDL and the 426 contacting veterinarian, making it possible to make new contacts and schedule visits to the properties. 427 This was demonstrated by the present study, in which users who did not send animals for necropsy or 428 tissue samples for histopathology in the first contact did so later, in addition to scheduling visits to the 429 properties where the problem occurred. Sending cadavers to the VDL for necropsy was considered an 430 important factor in reducing the number of inconclusive diagnoses, as this procedure is performed by 431 veterinarians trained for this practice (Pupin et al. 2019)

The contact made by field veterinarians located outside the LAP-UFMS area of operation, such as in the states of Mato Grosso, Goiás, Bahia and Pará, reinforces the usefulness of this remote tool for solving health problems since the rapid exchange of information and images from this means of communication help professionals to take certain actions for the definitive diagnosis, avoiding the loss of other animals due to diseases that can be controlled. The initial contact with VDLs located far from the property in question allows the team to indicate VDLs close to where the contacting veterinarian works.

438 The percentage of conclusive diagnoses in the necropsies performed at the LAP-UFMS was 71.11%. 439 This percentage is higher than previous studies, which accounted for 62.9% (Lucena et al. 2010) and 440 46.21% (Pupin et al. 2019), and in cases where veterinarians sent samples for histopathology. Although 441 the number of samples and the time interval evaluated in previous studies are higher than those considered 442 in this paper, this improvement in the diagnostic process can be attributed to the possibility of guiding 443 veterinarians in real time on the procedures to be adopted in the collection of samples during the 444 necropsy, according to the different clinical suspicions. This is reinforced by the good percentage of 445 conclusive diagnoses in samples sent by field veterinarians (79.5%), even when, in most cases (60%), the 446 initial clinical suspicion did not coincide with the final diagnosis. Even with a mistaken initial suspicion, the 447 exchange of information in real- time with members of the VDL team allowed the collection of information 448 and samples necessary for a conclusive diagnosis. Failure to send epidemiological data and anatomical 449 portions of organs essential for diagnosis and failures in packaging and conservation of samples were 450 the main reasons for inconclusive diagnoses.



485 486	Fig.1-4. Poisoning by <i>Brachiaria radicans</i> in cattle. (1) Good quality. Diffusely icteric carcass. The liver is diffusely dark brown (2) High quality. The kidney is diffusely black with a pelvis moderately vellowish (3) Good quality.
400	In the pacture, there are concerned of S and a concerned to (a) by the pacture theorem (a) and $($
488	predominates. The necropsy findings were compatible with intravascular hemolysis, and the absence of muscle
489	necrosis made it possible to exclude <i>S. occidentalis</i> intoxication from the differential diagnosis. The absence of
490	hemoparasites in the complementary exams and the spontaneous recovery of the cattle after the removal of the
491	paddock infested by <i>B. radicans</i> was the criterion for excluding the suspicion of babesiosis. Copper toxicosis was
492 493	excluded, as the cattle were raised extensively and received only mineral salt formulated for the affected animal category. Images provided by veterinarian Pedro Henrique Pereira de Jesus.



Fig.5-7. Poisoning by Stryphnodendron fissuratum in cattle raised extensively. (5) Good quality. Increased volume in the dewlap 532 region and skin dryness in the axillary and neck regions. (6) High quality. Subcutaneous in the dewlap region is thickened by yellowish gelatinous material (edema). (7) High quality. S. fissuratum fruit was found in the pasture where the cattle lived. In the smaller image, there are seeds of this plant, which were found in the rumen content of the necropsied bovine. The images are compatible with this intoxication despite not sending tissue samples for histopathological examination. Images 536 provided by veterinarian Fernando Arévalo Batista.

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538 An important example is the absence of spinal cord or brain samples for diagnosing rabies, the 539 main cause of death in cattle in this study and Mato Grosso do Sul (Ribas et al. 2013, Pupin et al. 2019). 540 As the correction of this failure represents an increase in conclusive diagnoses, communication between 541 the VDL team and the field veterinarian via a mobile application becomes important, as it is a fast way 542 that allows directing the professional in the collection of specific tissue samples according to clinical suspicion, and acts as a gateway for sending animals to the VDL for necropsy performed by thespecialized team.

These guidelines were fundamental for the efficiency of the diagnosis in this study since, in most cases, the initial suspicion did not correspond to the final diagnosis. This remote means of communication proved to be very useful in diseases that do not cause mortality, such as outbreaks of dewlap edema in cattle (Fig.10), a reaction possibly associated with hypersensitivity after drops in ambient temperature but can be confused with outbreaks of poisoning by toxic plants, such as Brachiaria sp. (Souza et al. 2010), Enterolobium contortisiliquum (Pupin et al. 2019) and Stryphnodendron fissuratum (Guizelini et al. 2021). In these situations, as tissue samples are often not sent to the VDL, the referral of images and clinical-epidemiological information is essential for the diagnosis.

Regarding the images that illustrate clinical signs and necropsy findings, the main limitation was a large number of images with poor quality, which reduced the chances of assistance to the veterinarian by the VDL team in the orientation for the accomplishment of the final diagnosis. Although the quality of the images was not ideal in many cases, the set of images and the clinical and epidemiological information provided helped formulate the conduct for the conclusive diagnosis. For this reason, the criterion adopted in the present study, which considered a conclusive diagnosis only in cases in which materials are sent for laboratory tests, is subject to review since, in several situations observed, the images associated with the history were sufficient for the formulation of the diagnosis.



Fig.8-9. Ruminal acidosis in feedlot cattle. (8) High quality. Rumen with extensive reddish areas and mucosal detachment. (9) Good quality. Rumen with extensive reddish areas. Although this organ was not referred for histopathological examination, the macroscopic diagnosis made it possible to take measures to contain the outbreak. In this case, there was no specific initial suspicion, but based on the clinical and epidemiological history, guidelines were developed on the conduct to be adopted during the necropsy. Images provided by veterinarian Helton Martins.



Fig.10. Edema in the dewlap region in cattle. High quality. The condition occurs as outbreaks in several rural properties and affects many cattle, usually after an intense cold. Recovery is spontaneous, but this condition must be distinguished from hepatogenous photosensitization caused by intoxication by *Brachiaria* spp., *Enterolobium contortisiliquum*, and *Stryphnodendron fissuratum*, in addition to heart failure and its causes. The initial suspicion of intoxication by *Brachiaria* sp. was ruled out based on clinical course and condition, with spontaneous recovery and without photodermatitis. Serum levels of the enzyme gamma-glutamyl transferase (GGT) and aspartate aminotransferase (AST) were within physiological values and were considered complementary diagnostic criteria. Image provided by the veterinarian Guilherme Dutra, Rodrigo Spengler and Luiz Carlos Louzada Ferreira.

640 Messaging applications for smartphones are useful tools to optimize and improve the efficiency of 641 diagnosis in the region where the VDLs operate, and, in this study, it helped to increase the number of 642 conclusive diagnoses in the LAP- UFMS due to the direct contact with field veterinarians during the 643 investigation of causes of death in farm animals. Results superior to those observed by our team can be 644 achieved if users of this type of service are trained for this purpose since the improvement in the quality of 645 the images and the clinical- epidemiological history help in the conclusion of the diagnosis at a distance, even 646 when samples are not forwarded to the VDL. Considering that flaws in the methodology for collecting and 647 sending tissue samples and pathological, clinical, and epidemiological data to the VDLs still exist, it is 648 necessary to develop training that approaches these aspects for field veterinarians. This approach may 649 not apply to countries that have a broad network of VDLs (Derscheid et al. 2021, Trevisan et al. 2021). However, 650 in countries with large territorial areas of animal production but do not have a sufficiently broad network 651 of VDLs, the search for alternatives that facilitate access to these VDLs is feasible to increase the diagnosis 652 of cattle diseases.

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CONCLUSION

Messaging applications for smartphones can improve the efficiency of diagnoses made by veterinary
 diagnostic laboratories (VDLs) and help field veterinarians in real time when collecting and sending
 samples.

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CAPÍTULO 3 – Cerebral Babesiosis: Transplacental Infection by Babesia bovis in a Calf

CASE REPORT Pub. 905

Cerebral Babesiosis: Transplacental Infection by Babesia bovis in a Calf

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763 ABSTRACT

Background: Cerebral babesiosis is a significant cause of mortality in calves, as it can be transmitted through the placenta during gestation and presents with hemolytic and/or neurological symptoms. However, the role of this agent as a cause of abortion is still poorly understood. In endemic areas, the disease primarily affects cattle between the ages of 1 and 12 months and occasionally neonates. This study aimed to describe the epidemiological, clinical, anatomopathological, molecular, and differential diagnostic aspects of a naturally infected newborn Nelore calf.

Case: A newborn Nelore calf from Mato Grosso do Sul was discovered dead 1 hour 771 772 after birth and was subjected to necropsy, cytopathological and histopathological examinations by the Pathological Anatomy Laboratory (LAP) at the Federal University of 773 774 Mato Grosso do Sul (UFMS), as well as complementary in situ hybridization (ISH) and polymerase chain reaction (PCR) tests. Hemoparasite research yielded negative results in all 775 cows within the herd, including the mother of the necropsied calf, which were all clinically 776 777 healthy. The clinical and hematological examination results of the cows were normal. At necropsy, the calf appeared thin, with a moderately jaundiced carcass and multiple petechiae 778 on the pleural and pericardial surfaces. The lung was inflated until the opening of the thoracic 779 780 cavity and floated when immersed in formalin, indicating that the calf was born alive and

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781 breathing. The spleen and liver were moderately enlarged with rounded edges, and the liver appeared slightly orange. The gray matter of the brain was significantly cherry red. 782 783 Histologically, the central nervous system capillaries were slightly congested, with numerous punctiform and basophilic structures observed in the erythrocytes obliterating these vessels, 784 785 either alone or in pairs, measuring 1 to $2 \mu m$ in diameter, which were morphologically consistent with B. bovis. In the cytological evaluation of the imprint slides of the brain cortex, 786 blood capillaries filled with erythrocytes parasitized by solitary or paired punctiform 787 788 basophilic structures, approximately 1 µm in diameter, morphologically compatible with B. 789 bovis, were detected. Positive labeling for Babesia spp. was observed in the ISH examination, while the PCR identified B. bovis and B. bigemina in were identified in the 790 791 brain fragments. The macroscopic findings, including pale or icteric mucous membranes, yellow and enlarged liver, and splenomegaly observed in cases of anaplasmosis, may be 792 confused with those of cerebral babesiosis when the latter does not present with 793 hemoglobinuria. Brain congestion indicates B. bovis infection but does not rule out co-794 795 infection with Anaplasma marginale in this case, although this rickettsia was not identified 796 in the blood smear.

797 Discussion: The diagnosis of B. bovis infection was based on epidemiological and 798 histological data, as well as the identification of the protozoan in spleen and brain smears 799 and through PCR and ISH exams, which are highly sensitive and can aid in the diagnosis of 800 B. bovis in cases of perinatal deaths. In situ hybridization is effective in cases where the 801 material has autolysis, as the technique allows the genetic material of the agent to be 802 associated with the lesion, even with tissue alterations caused by fixation in formalin. The 803 findings of this study highlight the importance of considering this disease as a differential diagnosis among those that cause abortions or neonatal losses in cattle and emphasize the 804 805 importance of conducting anatomopathological exams for definitive diagnosis.

Keywords: Cerebral babesiosis, *Babesia bovis*, pathology, transplacental
transmission, intrauterine infection, *in situ* hybridization.

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INTRODUCTION

Cerebral babesiosis, caused by the protozoan *Babesia bovis*, is a significant cause of death in cattle, presenting with hemolytic and neurological symptoms [5,8,30]. The protozoan is transmitted by the tick *Rhipicephalus microplus*, which is distributed in tropical and subtropical areas, making babesiosis a condition of enzootic stability in these regions [25,26,28].

The occurrence of the disease and deaths caused by it are influenced by various epidemiological factors, including the transmission capacity of the tick population, as well as the susceptibility of cattle related to their immunological status, breed, and age of affected animals [10,25,30]. The decline in colostral antibody levels and the high burden of tick infestation, leading to the phase of greatest susceptibility to infection, can result in the development of the clinical manifestation of the disease, with high rates of morbidity and mortality [10].

Considering that babesiosis is a disease that has been causing significant neonatal losses and abortions in cattle [1,14,15,20], it is important to consider this disease as a differential diagnosis for babesiosis caused by *B. bigemina* and anaplasmosis [3,4,9,12], as well as for bovine infectious rhinotracheitis (IBR), bovine viral diarrhea (BVDV), leptospirosis, neosporosis, and brucellosis [19]. However, reports of neonatal deaths and transplacental infections caused by *Babesia* sp. are rare [30].

This study aimed to describe a case of bovine neonatal death caused by *B. bovis* in the state of Mato Grosso do Sul, covering the main epidemiological, clinical, pathological, and molecular aspects, as well as the differential diagnoses for this disease.

830

CASE

A newly born Nellore calf underwent necropsy and histopathological examination by 832 the team at the Pathological Anatomy Laboratory (LAP) of the Federal UFMS. 833 834 Epidemiological and clinical data of the affected herd were obtained through an interview with the veterinarian responsible for the herd during visits to the property by the LAP-UFMS 835 team where the case occurred. During the visits, all cows were subjected to body temperature 836 837 measurement, and blood samples were collected by jugular vein puncture, which was stored 838 in tubes containing EDTA anticoagulant for direct investigation of hemoparasites and a complete blood count. 839 840 In April 2019, a male Nelore calf was born on a rural property in Maracaju (21° 36' 841 52" S; 55° 10' 06" W), Mato Grosso do Sul state, from a healthy cow and was found dead one hour after birth. The property where the case occurred consisted of 18 dairy cows raised 842 843 extensively on pasture under tropical climate conditions. During a visit to the property, it 844 was found that the cows had an intense infestation of R. microplus ticks during the last months of gestation. 845 846 During the necropsy, fragments from various organs were collected, fixed in a 10 % formalin solution, routinely processed for hematoxylin and eosin (HE)¹ staining, and 847 examined under light microscopy. Additional procedures were carried out to investigate 848 849 hemoparasites, such as spleen imprints and cerebral cortex smears stained with Panótico

850 Rápido \mathbb{R}^1 .

Brain fragments were collected, refrigerated, and sent for PCR tests. Paraffinembedded brain blocks were sent for *in situ* hybridization (ISH). PCR used the Cybi/Cbbr² primers specific for *B. bigemina* and Cybo/Cbbr² primers specific for *B. bovis* [6]. *In situ* hybridization procedures followed the methodology described in 2020 by Hülskötter *et al.* [16]. The previously used riboprobe (probe for ribosomal RNA) detected *Babesia microti* in experimentally infected chickens. The primers for *B. microti* 16S rRNA similar gene were
BM16SFW 5'-CAT GTCTTAGTA TAA GCT TTT ATA CAG CGA AA-3' and BM16SRW
5'-AAC GCT CGG AAG CGA GAT TAA TGA CAA GGC AG-3'. As negative controls for
the technique, chicken tissues infected with a probe in the same direction were processed.

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RESULTS

The results of the hemoparasite research were negative in all cows in the herd, 861 862 including the necropsied calf's mother, that was heavily parasitized by ticks. The clinical and 863 hematological examination results of the cows did not show abnormalities. At necropsy, the 864 calf was thin, with a moderately icteric carcass and multiple petechiae on the pleural and pericardial surfaces. The lung was armed to the opening of the thoracic cavity and floated 865 866 when immersed in formaldehyde, indicating that there was breathing and, therefore, the calf was born alive. There was no colostrum in the abomasum, indicating that no colostrum had 867 been fed. The spleen and liver were diffusely and moderately enlarged with rounded edges. 868 The liver was slightly orange. The brain's gray matter was markedly cherry red (Figure 1). 869

Histologically, the capillaries of the central nervous system were diffusely and mildly 870 871 congested; numerous pinpoint and basophilic structures measuring 1-2 µm in diameter, morphologically compatible with B. bovis, were observed in the red blood cells that 872 obliterated these vessels. Cytological evaluation of the imprint slides of the cerebral cortex 873 874 showed blood capillaries filled with erythrocytes parasitized by solitary or paired basophilic 875 pinpoint structures, approximately 1µm in diameter, morphologically compatible with B. 876 bovis (Figure 2A). ISH examination showed positive labeling for *Babesia* spp. (Figure 2B). 877 The PCR analysis identified *B. bovis* and *B. bigemina* in the brain fragments.

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DISCUSSION

879 The diagnosis of *B. bovis* infection as the cause of death in this calf was based on 880 epidemiological data, macroscopic and histological findings, and the identification of the protozoan in spleen and brain imprints, as well as through PCR and ISH. Similar findings
have been described in aborted fetuses, stillborns and neonates resulting from transplacental *B. bovis* infection in cattle [14,21,24].



Figure 1. Macroscopic changes in a calf infected *in utero* by *Babesia bovis*. Transverse
section of the frontal portion of the brain. The gray matter is diffusely cherry red.

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Previous reports of abortions or neonatal deaths caused by *B. bovis* have shown that mothers were asymptomatic at the time of parturition [9,14,29], similar to the present case where all cows in the herd were clinically healthy, and *Babesia* spp. was not detected in blood smears, including that of the cow that gave birth to the necropsied calf. This occurs because adult animals living in areas of enzootic stability have sufficient immunity and normally do not become ill, and tests such as blood smears or complete blood counts do not detect minimum levels of infection [9].

903 The pathogenesis of transplacental infection by *B. bovis* is not well understood. 904 However, some authors suggest that the transmission capacity may be affected by the 905 intrinsic characteristics of the parasite, such as size, shape, and strain, and of the host, such 906 as damage to blood vessels in the placental membrane and immunity [9]. Another hypothesis is that synepitheliochorial placentation in ruminants allows fetal infection by pathogenic 907 908 agents but limits the transfer of larger molecules, mainly antibodies, from mother to fetus [14,28,29]. Although it does not show clinical signs of the disease and no hemoparasites 909 910 were found, the calf's mother was heavily parasitized by ticks. It is possible that the cow had 911 the presence of the protozoan at basal levels and transmitted it to the calf through the 912 placenta, this happens in chronically infected cows living in areas of enzootic stability, as 913 observed in some studies [17].



Figure 2. Cytological and molecular findings in a calf infected *in utero* by *Babesia bovis*. A- Cytology of the cerebral cortex smear. Blood capillaries are filled with erythrocytes
containing basophilic structures measuring approximately 1-2 µm in diameter, compatible
with *Babesia bovis* [Panotic Rapid; 10x]. B- Brain. Positive staining for *Babesia* sp. inside
erythrocytes [*In situ* hybridization; 40x].

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Tick infestation in pregnant cows may be a result of immunosuppression that occurs two to three weeks before calving. [18]. In one study, a greater demand for oxygen by the fetus was observed in the last third of pregnancy, which may be one of the determinants of fetal death in cases of lack of blood supply [23]. Therefore, it is possible that the reduction in blood supply could lead to fetal anoxia and its consequent death.

Besides babesioses, other diseases may cause abortions in cattle, including 931 anaplasmosis, leptospirosis, and neosporosis [2,7,9,11,13,14,22]. In anaplasmosis, the 932 933 macroscopic findings, including pale or icteric mucous membranes, yellow and enlarged liver, and splenomegaly in anaplasmosis, can be confused with those of cerebral babesiosis 934 935 when it does not present hemoglobinuria [9]. In our case, brain congestion demonstrates 936 infection by B. bovis but does not exclude co-infection by Anaplasma marginale. However, this rickettsia was not identified in the blood smear. Infection by N. caninum is an important 937 938 cause of abortion in cattle, and post-mortem macroscopic findings include degenerative to 939 inflammatory lesions of fetal tissues. However, in this case no macroscopic changes were observed.. 940

941 Both PCR and ISH confirmed the presence of *B. bovis* in the necropsied calf; however, its high sensitivity should be interpreted cautiously. As babesiosis is confirmed by 942 visualizing the protozoan in red blood cells, the use of molecular techniques (PCR and ISH) 943 to identify the pathogen associated with necropsy and histological findings facilitates the 944 945 confirmation of the diagnosis, even in cases where the smear is not performed or the parasite 946 is not visualized [6,16]. In cases of autolysis, these techniques are also useful, as ISH mainly 947 associates the genetic material of the agent with the lesion. The fact that B. bigemina was identified in the brain tissue does not indicate that this agent was responsible for the death of 948 949 the calf, it only points to the presence of infection. Macroscopic and microscopic findings 950 demonstrated the characteristic brain lesions caused by *B. bovis* would not be observed in *B.* 951 bigemina [26].

This case demonstrates the need to include cerebral babesiosis in the list of differential diagnoses of diseases that cause neonatal deaths in cattle. Therefore, the importance of conducting necropsy exams, smears of fragments of the cerebral cortex, and

955	histopathological exams to reach a conclusive diagnosis is highlighted. Identifying the agent
956	through PCR and ISH proved to be useful tools for the conclusion of the diagnosis.
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961	authors are responsible for the content and writing of this article.
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CAPÍTULO 4 - The use of WhatsApp to identify previously undiagnosed and underreported ruminant and equine diseases in Mato Grosso do Sul

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ABSTRACT. - Bonato G.C., Souza L.L., Silva T.X., Pupin R.C., Gomes D.C., Araújo F.S. & Lemos R.A.A.
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1075 This study explores the use of WhatsApp as a tool for identifying previously undiagnosed and 1076 underreported diseases in ruminants and horses in Mato Grosso do Sul, Brazil. The research, carried out 1077 from January 2022 to December 2023, involved a WhatsApp group made up of 302 public service and 1078 private sector veterinarians. The contacts were categorized as those that resulted in a necropsy by the 1079 veterinary diagnostic laboratory, the sending of a sample for histopathological analysis, or the non-1080 sending of material. The results demonstrate the efficiency of using WhatsApp for diagnosing 1081 underreported diseases such as anaplasmosis, haemonchosis, rabies, blackleg disease, Mannheimia 1082 haemolytica pneumonia, bovine herpesvirus encephalitis, multiple mineral deficiency in cattle and 1083 copper poisoning in sheep, thus increasing diagnostic efficiency for veterinary laboratories. The tool also 1084 facilitated communication, resulting in an increase in conclusive diagnoses, the identification of 1085 previously undiagnosed diseases such as Chlamydia pecorum encephalomyelitis in buffalo, peripheral 1086 neuropathy in cattle, phosphorus deficiency in cattle, vitamin A deficiency, and perinatal pneumonia caused by Salmonella enterica subsp. Arizonae in cattle. In addition, it was possible to identify 1087 1088 underreported occurrences such as cases of abortion and neonatal diarrhea and to implement the 1089 correction of diagnostic approaches in real time via WhatsApp. The study concludes that WhatsApp 1090 improves communication between field veterinarians and diagnostic laboratories, providing valuable 1091 information on disease occurrence and diagnostic challenges.

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INDEX TERMS: Cattle, diagnostic laboratories, underreported diseases, WhatsApp.

1095 RESUMO: Este estudo explora o uso do WhatsApp como ferramenta para identificação de doenças até então não diagnosticadas e subnotificadas em ruminantes e equinos em Mato Grosso do Sul, Brasil. A 1096 1097 pesquisa, realizada de janeiro de 2022 a dezembro de 2023, envolveu um grupo de WhatsApp composto por 302 veterinários do serviço público e da iniciativa privada. Os contatos foram categorizados naqueles 1098 1099 que resultaram em necropsia pelo laboratório de diagnóstico veterinário, ao envio de amostra para 1100 análise histopatológica ou ao não envio de material. Os resultados demonstram a eficiência do uso do 1101 WhatsApp para o diagnóstico de doenças subnotificadas como a anaplasmose, hemoncose, raiva, 1102 carbúnculo sintomático, pneumonia por *Mannheimia haemolytica*, encefalite por herpesvírus bovino, 1103 deficiência mineral múltipla em bovinos e intoxicação por cobre em ovinos, ampliando assim a eficiência 1104 diagnóstica para os laboratórios veterinários. A ferramenta também facilitou a comunicação, resultando 1105 em aumento de diagnósticos conclusivos, identificação de doenças não diagnosticadas previamente 1106 como a encefalomielite por Chlamydia pecorum em búfalos, neuropatia periférica em bovinos, deficiência 1107 de fósforo em bovinos, deficiência de vitamina A e a pneumonia perinatal por *Salmonella enterica* subsp. 1108 Arizonae em bovinos. Além disso, foi possível identificar ocorrências subnotificadas como casos de 1109 aborto e diarreia neonatal, e implementar a correção de abordagens diagnósticas em tempo real via 1110 WhatsApp. O estudo conclui que o WhatsApp melhora a comunicação entre veterinários de campo e 1111 laboratórios de diagnóstico, fornecendo informações valiosas sobre a ocorrência de doenças e desafios 1112 de diagnóstico.

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TERMOS DE INDEXAÇÃO: Gado, laboratórios de diagnóstico, doenças subnotificadas, WhatsApp.

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1117 Veterinary diagnostic laboratories are fundamental components of an integrated animal health 1118 system and dedicated to the study of diseases that cause losses or limit animal production. In addition to 1119 diagnosis, their work aims to determine the conditions in which diseases occur, enabling the adoption of 1120 efficient measures to control them (Lemos et al. 2023). Their actions are responsible for a significant 1121 reduction in economic losses caused by diseases (Schulz et al. 2018). Despite their importance, factors 1122 limiting the efficiency of veterinary diagnostic laboratories have been identified in various studies, such 1123 as the sending of inadequate materials for diagnosis and the lack of epidemiological and clinical 1124 information regarding the materials sent (Lucena et al. 2010, Rondelli et al. 2017, Pupin et al. 2019).

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1125 The use of the instant messaging application for smartphones, WhatsApp, as a tool to reduce 1126 these limitations has proven to be efficient, considerably reducing the number of inconclusive diagnoses 1127 (Lemos et al. 2023). However, the use of this tool revealed cases in which, despite contact between 1128 veterinarians working in the field and the veterinary diagnostic laboratory, no material was sent, which 1129 results in the underreporting of diseases when studies are carried out only on the basis of samples sent 1130 to laboratories. In addition, diseases that had not been diagnosed in other years in the region were 1131 diagnosed after using this tool. 1132 In this study, we evaluated the use of the WhatsApp application as a tool for diagnosing

In this study, we evaluated the use of the WhatsApp application as a tool for diagnosing previously undiagnosed and underreported diseases in ruminants and horses in Mato Grosso do Sul.

METHODOLOGY

Etthical Approval. This study was not submitted to the Ethics Committee on the Use of Animals (CEUA) of the "Universidade Federal de Mato Grosso do Sul" (UFMS), because the cases studied were sick or dead cattle from laboratory routine and the legislation only provides for the use of live animals for experimentation.

The data were collected from a WhatsApp group made up of 302 veterinarians from the public and private sector, who were assessed by the team at the Pathology Laboratory at the Federal University of Mato Grosso do Sul (LAP-UFMS). The collection period was from January 2022 to December 2023, totaling 24 months.

The WhatsApp contacts were divided into three categories: (1) contact that resulted in the animal being sent for necropsy or where the necropsy was carried out on the property by the LAP-UFMS team, (2) contact that resulted in animal tissue samples being sent to the LAP-UFMS, collected by the group's veterinarians, for histopathological analysis, and (3) contacts with no animal sent to necropsy or material for histopathological evaluation. In the first two situations, contacts were made after the cases had been notified, in other words, to obtain further information until the outbreak was closed.

In cases where samples were not sent to the LAP-UFMS, the methods used to obtain diagnostic suspicion or conclusive diagnosis were divided into three parts: 1 – collection of samples for other laboratory tests; 2 – images or video sent; 3 – positive response to the treatment instituted.

For analysis purposes, cases that were related to each other and occurred on the same property were grouped together as a single contact, regardless of the number of necropsies performed.

RESULTS

From January 2022 to December 2023, 63 contacts were obtained that resulted in necropsies carried out by the LAP-UFMS team, 62 samples were received (including biopsies and necropsy materials), and 86 contacts were made without sending material, all using WhatsApp to communicate and/or send.

The frequencies of occurrences diagnosed in ruminants and horses through WhatsApp contacts that resulted in necropsies by the LAP-UFMS are shown in Table 1.

The diagnoses made using the samples sent for histopathological evaluation by veterinarians outside the LAP-UFMS are listed with their respective frequencies in Table 2.

Of the total of 125 cases analyzed during the study period, 63 of the necropsies carried out by the LAP-UFMS team and 62 of the samples sent by veterinarians outside the LAP for histopathology, 21 cases (16.8%) resulted in inconclusive diagnoses.

1168The diagnoses or suspected diagnoses of LAP obtained through contacts that did not result in1169animals being sent for necropsy or materials for histopathological examination are listed in Table 3 with1170the applicants' complaints, the diagnostic methods used, and their frequency of occurrence.

1171 In addition to the 29 cases in which no samples were sent for histopathology or necropsy but 1172 which were suspected or diagnosed using LAP-UFMS methods, in 57 cases, there was only clinical 1173 information about what was happening or the veterinarian's suspicion. Therefore, it was not possible to 1174 elucidate the cause of clinical signs and mortalities. DISCUSSION WhatsApp has proven to be an efficient tool for identifying underreported diseases in production animals. The use of the tool to increase the efficiency of Veterinary Diagnostic Laboratories (VDLs) services has been demonstrated previously (Lemos et al. 2023), but its use for identifying underreported diseases has not been evaluated. Its use for correcting diagnostic procedures has also not been evaluated. Previously undiagnosed diseases. Among the diseases not previously diagnosed at LAP-UFMS are encephalomyelitis caused by Chlamydia pecorum in buffaloes, peripheral neuropathy in cattle, phosphorus deficiency in cattle, and vitamin A deficiency and perinatal pneumonia caused by Salmonella enterica subsp. Arizonae in cattle. WhatsApp was an essential tool for the diagnosis of the first two as the diagnosis was not made by the first necropsies, and monitoring the continuity of the outbreaks through contact between producers and veterinarians responsible for these cases made it possible to correct the diagnostic approach. Chlamydia pecorum encephalomyelitis has not previously been described in ruminants in Brazil, and cases in buffaloes are rare worldwide (Astarita et al. 2007, Rigamonti et al. 2022). Peripheral neuropathy is also not reported in cattle, although a similar disease has been described in horses in other countries. (Furuoka et al. 1994, 1999, Hanche-Olsen et al. 2008, Hanche-Olsen et al. 2017). Clinically, this disease is characterized by digital extensor dysfunction, mainly affecting the pelvic limbs with the animal remaining alert, without presenting other clinical manifestations (Figure 1). In these cases, the only lesion observed was degeneration of the peripheral nerves, which corroborated the diagnosis.

Table 1. Diagnoses in ruminants and horses obtained through necropsies carried out by the LAP-UFMSteam, from January 2022 to December 2023.

Diagnosis	Frequency
Cattle	
Rabies	19.04% (12/63)
+Anaplasmosis	14.28% (9/63)
Blackleg disease	4.76% (3/63)
Inconclusive	4.76% (3/63)
*Perinatal deaths	66.6% (2/3)
Bovine herpesvirus meningoencephalomyelitis	6.34% (4/63)
Vitamin E and selenium deficiency	3.17% (2/63)
Enzootic bovine leukosis	3.17% (2/63)
Septicemic salmonellosis e Anaplasma marginale	3.17% (2/63)
Botulism	1.58% (1/63)
Malignant catarrhal fever	1.58% (1/63)
Hypocalcemia	1.58% (1/63)
Cobalt deficiency	1.58% (1/63)
Phosphorus deficiency	1.58% (1/63)
Sodium deficiency	1.58% (1/63)
Vitamin A deficiency	1.58% (1/63)
Hypothermia	1.58% (1/63)
Sodium poisoning	1.58% (1/63)
Stryphnodendron fissuratum poisoning	1.58% (1/63)
Multicentric juvenile leukosis	1.58% (1/63)
Multiple Malformations	1.58% (1/63)
Peripheral neuropathy	1.58% (1/63)
Osteomyelitis due to spinal cord compression	1.58% (1/63)
Papillomatosis	1.58% (1/63)
Peritonitis	1.58% (1/63)
Polioencephalomalacia	1.58% (1/63)
Mannheimia haemolytica pneumonia	1.58% (1/63)
Perinatal pneumonia by <i>Salmonella enterica</i> subsp. Arizonae	1.58% (1/63)
Suggestive of poisoning by Microcystis aeruginosa	1.58% (1/63)
Bubaline	
Encephalomyelitis caused by Chlamydia pecorum	1.58% (1/63)
Equine	
Secondary nutritional hyperparathyroidism	1.58% (1/63)
Suggestive of encephalomyelitis by alfavírus	1.58% (1/63)
Cecocolic intussusception and peritonitis	1.58% (1/63)
Snakebite envenoming by Bothrops spp.	1.58% (1/63)
Ionophore antibiotic poisoning	1.58% (1/63)
Grand Total	100.00% (63)

*Most frequent among the inconclusive cases; +Diagnosis of haemonchosis associated with one
 of the cases

1238 Regarding phosphorus deficiency, its occurrence in soils and pastures is described in several 1239 Brazilian states, including Mato Grosso do Sul (Tokarnia et al. 2000). Although it is considered the main mineral deficiency in Brazil, the description of outbreaks of diseases with clinical and pathological 1240 1241 follow-up is scarce in ruminants (Malafaia et al. 2023) and has not been previously reported in Mato 1242 Grosso do Sul (Pupin et al. 2019). In these cases, epidemiological, clinical, and pathological 1243 characterization, as well as monitoring the response to treatment, are facilitated with the use of 1244 WhatsApp. In the case diagnosed, it was possible to observe the occurrence of spontaneous fractures in 1245 the animals and the fragility of multiple bones, in addition to inadequate mineral supplementation 1246 (Figures 2–3).

1247 The diagnosis of vitamin A deficiency was also made possible by monitoring the evolution of the 1248 outbreak. Initially, the suspicion was polioencephalomalacia based on the blindness presented by the 1249 cattle, but as there was no regression of this clinical sign with treatment or evolution to other 1250 neurological signs, the case was reported to the LAP-UFMS via the app and diagnosed for the first time 1251 in Brazil (Pupin et al. 2023), with no other descriptions found in the literature.

1252 Another diagnosis obtained in the present study that has not been described in previous studies 1253 is perinatal pneumonia caused by Salmonella enterica subsp. Arizonae, which refers to a case of perinatal 1254 death of a calf that had been born weak, through normal birth, and did not get up or suckle. On 1255 histopathology, the calf presented marked interstitial pneumonia with foamy macrophages and a 1256 predominance of neutrophils, with intralesional agents. In complementary lung exams, there was growth in the bacterial culture of Salmonella enterica subsp. Arizonae and positive immunohistochemical 1257 1258 staining for the genus Salmonella. Cases of salmonellosis in calves described in MS or other regions 1259 presented mononuclear interstitial pneumonia, associated or not with intestinal lesions, in calves over 1260 30 days of age, caused by Salmonella enterica subsp. enterica serotype Dublin (Guizelini et al. 2019, 2020). 1261 Both the characteristics of the histological lesion, the age of the affected calf and the bacterial agent 1262 isolated in the culture, differ from typical cases of salmonellosis in calves. Salmonella Dublin is considered a cause of abortion and stillbirth (Sanchez et al. 2018, Mee. 2023), and according to the 1263 1264 incubation period of the disease, this case suggests that it is an intrauterine infection.

1265 Diseases with changed conditions of occurrence. Some diseases have already been diagnosed 1266 in Mato Grosso do Sul, but their conditions of occurrence have changed. With the increase in the area 1267 devoted to agriculture and the planting of trees for cellulose production, there has been a significant reduction in the area devoted to pasture (McManus et al. 2016), resulting in an intensification of cattle 1268 1269 production with the increase in crop-livestock integration systems and feedlots. The main consequence 1270 of this change is the concentration of herds destined for calving in the Pantanal region and that of 1271 finishing herds in the plateau region. As a result, diseases that previously occurred mainly in extensive 1272 production systems began to be diagnosed more frequently in these other farming systems. This was 1273 evidenced in this study with anaplasmosis, cobalt deficiency, sodium deficiency, hypothermia, 1274 haemonchosis, and dictyocaulosis. 1275

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Diagnosis	Frequency
Cattle	
Inconclusive	24.19% (15/62)
Rabies	16.12% (10/62)
Babesiosis	8.06% (5/62)
Bovine herpesvirus meningoencephalomyelitis	6.45% (4/62)
Hypothermia	4.83% (3/62)
Polioencephalomalacia	3.22% (2/62)
Mannheimia haemolytica pneumonia	3.22% (2/62)
Septicemic salmonellosis	3.22% (2/62)
Anaplasmosis	3.44% (3/62)
Blackleg disease	3.22% (2/62)
Bacterial abscesses	1.62% (1/62)
Lick dermatitis	1.62% (1/62)
Pododermatitis	1.62% (1/62)
Necrohemorrhagic abomasitis	1.62% (1/62)
Brachiaria sp. poisoning	1.62% (1/62)
Eimeriosis	1.62% (1/62)
Equine	
Inconclusive	4.83% (3/62)
Secondary nutritional hyperparathyroidism	3.22% (2/62)
Clostridial myositis	1.62% (1/62)
Sarcoid	1.62% (1/62)
Rabies	1.62% (1/62)
Grand Total	100.00% (62)

Table 2. Diagnoses in ruminants and horses obtained through histopathological samples sent by
 veterinarians external to LAP, from January 2022 to December 2023.

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Outbreaks of anaplasmosis affecting a significant number of cattle were diagnosed in integrated
 crop-livestock systems, feedlots, and extensive production systems. In all cases, the cattle came from
 properties located in the Pantanal and were introduced into properties located on the plateau. This is a
 condition considered a predisposing factor for the occurrence of the disease (Puentes & Riet-Correa,
 2023), but its occurrence in different production systems has not been recorded previously.

Cobalt deficiency has previously been described in the state in extensive systems (Silva et al. 2020). In the present study, however, the disease occurred in a crop-livestock integration system, and some differences between the two systems, related to the disease, deserve to be highlighted. In integration systems, the pasture is fertilized, which reduces the need for phosphorus supplementation. However, cobalt is not essential for plants and is therefore not used in fertilization. In addition, the large amount of forage produced means that a large number of cattle can be brought onto the pastures. Therefore, mineral supplementation must be carried out properly, paying particular attention to the number of troughs available to the animals.

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Figure 1. Peripheral neuropathy in cattle. Complete flexion of the metatarsophalangeal joint of a bovine in a quadrupedal position in the early stages of the disease.

Table 3. Diagnoses or suspected diagnoses obtained through contacts without sending the animal fornecropsy or material to the LAP-UFMS for histopathological examination, from January 2022 to December2023.

Diagnoses or suspected diagnoses for LAP-UFMS	Applicant's complaint/suspicion	Method diagnosis	Frequency
Cattle			
Dewlap edema	Dewlap edema or not informed	2	8.13% (7/86)
Anaplasmosis	Pale mucous membranes, getting tired easily, lying down, losing weight, not eating, or not being informed	1;3	2.32% (2/86)
Dictiocaulose	Coughing	2;1	2.32% (2/86)
Hemoncosis	Not informed	1	2.32% (2/86)
Hepatogenic photosensitization	Dewlap edema or not informed	2	2.32% (2/86)
Babesiosis	Mortality, cattle with jaundice, splenomegaly, and dark urine	2;3	1.16% (1/86)
Bovine keratoconjunctivitis	Not informed	2	1,16% (1/86)
Cobalt deficiency	Not informed	1	1.16% (1/86)

Multiple mineral deficiency	Not informed	3	1.16% (1/86)
Dermatomycosis	Not informed	2	1.16% (1/86)
Hypothermia	Not informed	2	1.16% (1/86)
Stryphnodendron fissuratum poisoning	Mortality, neurological signs, and abortions	2	1.16% (1/86)
Mesothelioma	Not informed	2	1.16% (1/86)
Pododermatitis	Not informed	2	1.16% (1/86)
Polioencephalomalacia	Partial blindness	3	1.16% (1/86)
Parasitic otitis	Firm ear	2	1.16% (1/86)
Sheep			1.16% (1/86)
Copper poisoning	Not informed	2;1;3	1.16% (1/86)
Suggestive of copper poisoning	Not informed	2	1.16% (1/86)
Total			33.72% (29/86)

1- Other complementary exams; 2- sending photos or videos; 3- positive response to treatment

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Sodium deficiency has not been described in Mato Grosso do Sul, as shown by a 24-year survey 1341 of diagnoses made at LAP-UFMS (Pupin et al. 2019). However, after using WhatsApp in a more recent 1342 study, an outbreak was described in cattle raised extensively (Lemos et al. 2023). In the present study, 1343 the deficiency was diagnosed again in two outbreaks, both in feedlot cattle. At necropsy, this deficiency 1344 does not cause characteristic lesions (Barbosa et al. 2021), and the finding of piloconcrements in the 1345 rumen is the main evidence, making the inspection of this organ at necropsy and the interpretation of 1346 the finding associated with the clinical picture fundamental for the diagnosis. In this sense, real-time 1347 communication between the laboratory and the veterinarian working in the field can contribute to the 1348 diagnosis.

1349 Haemonchosis and dictyocaulosis in cattle have also been observed in integrated crop-livestock 1350 systems, with the case of haemonchosis diagnosed as a necropsy finding when attending to cases of mortality due to anaplasmosis. Given the need to acquire numerous cattle from different origins, which 1351 1352 are certainly subject to different health protocols, it is believed that many of these cattle are resistant to 1353 the active principles used on the destination property. This situation is common to both infections and was observed in this study. One particularity regarding dictyocaulosis is that in integrated crop-livestock 1354 1355 systems, there is often a lack of trees that could act as shelter and offer thermal comfort to the animals. This lack of shelter during periods of low temperatures favors the occurrence of parasitic pneumonia as 1356 1357 exposure to cold is a risk factor for the disease. Regarding the other cases of haemonchosis, these 1358 occurred in extensive production systems and were not the result of necropsies but rather those of complementary tests on live animals. The haemonchosis outbreaks previously described in the state 1359 were diagnosed only in cases of mortalities (Pupin et al. 2019). The occurrence of haemonchosis may 1360 1361 have increased since the deworming of cattle herds used to be carried out at the time of vaccination against foot-and-mouth disease in the state, and with the removal of mandatory vaccination, this practice 1362 1363 may be neglected.

1364 Although previously described in Mato Grosso do Sul (Santos et al. 2012), hypothermia occurred 1365 in places where the disease did not historically occur. Some of the outbreaks in 2023 are related to cattle 1366 transported from the Pantanal region after weaning and introduced into crop-livestock integration 1367 systems. In this situation, low body weight and the absence of trees are predisposing factors for
1368 outbreaks (Lemos et al. 2023).
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Table 4. Diagnostic suspicions or clinical information from veterinarians in the group that did not result in
 suspicion or diagnosis by the LAP-UFMS due to lack or scarcity of information, from January 2022 to
 December 2023.

Diagnosis suspicious/veterinarian's complaint	Frequency
Mortality	11.62% (10/86)
Neurological signs	10.46% (9/86)
Urea poisoning	4.65% (4/86)
Rabies	4.65% (4/86)
Pneumonia	3.48% (3/86)
Polioencephalomalacia	3.48% (3/86)
Breathing difficulty	2.32% (2/86)
Coughing	2.32% (2/86)
Skin lesions	2.32% (2/86)
Botulism	2.32% (2/86)
Blackleg disease	2.32% (2/86)
Abortion	2.32% (2/86)
Diarrhea in calves	2.32% (2/86)
Calves eat dirt and have white diarrhea	2.32% (2/86)
Yellowish diarrhea and calf mortality	1.16% (1/86)
Late-term abortions	1.16% (1/86)
Babesiosis	1.16% (1/86)
Cattle with tremors and in recumbency	1.16% (1/86)
Clostridiosis	1.16% (1/86)

Total	66.27% (57/86)
Plant poisoning	1.16% (1/86)
Cypermethrin and chlorpyrifos <i>pour-on</i> poisoning	1.16% (1/86)
Bovine herpesvirus	1.16% (1/86)



Figures 2–3. Clinical and epidemiological findings of phosphorus deficiency in cattle. 2) Dislocation of the right scapula and lack of support on the floor of the thoracic limb on the corresponding side in cattle with humeral fracture. 3) Broken troughs in a batch with an excessive number of cattle.

Monitoring of outbreaks and underreporting of diseases. In several previously diagnosed diseases, monitoring the evolution of outbreaks has shown that the real number of cases is higher than that recorded when the material is sent to the laboratory or when the first contact is made. This situation was observed mainly in outbreaks of anaplasmosis, rabies, blackleg disease, *Mannheimia haemolytica* pneumonia, and bovine herpesvirus (BoHV) encephalitis. In rabies outbreaks, in addition to underreporting, several cases were reported in feedlots, a situation previously not observed in the state (Ribas et al. 2013, Pupin et al. 2019). Underreporting of rabies has been described previously (Mello et al. 2019). When monitoring BoHV encephalitis outbreaks, in addition to the repetition of outbreaks on the same property, it was possible to observe affected animals aged less than 6 months and more than 48 months, which is considered rare in BoHV-5 infection (Salvador et al. 1998, Gomes et al. 2002, Rissi et al. 2007, Ribas et al. 2013).

Suggestive diagnoses. Among the diseases monitored by the LAP-UFMS team that did not have a conclusive etiological diagnosis are hypocalcemia in pregnant cows, Microcystis aeruginosa poisoning, and equine viral encephalomyelitis. The first situation was observed in a Nelore cow which was found lying down with a normal response to visual stimuli, sensitivity in the limbs, decreased mandibular and tongue tone, abdominal breathing with short movements, and congested ocular and vulvar mucous membranes. She was raised on Brachiaria brizantha pasture and was not recently calved. No significant macro- or microscopic lesions were observed. However, biochemical examination showed low serum calcium levels. Hypocalcemia in cattle is a disease that typically occurs in peripartum cows, but sporadic cases can occur when the cow is in another physiological stage and consumes pasture with a high oxalate content (Constable et al. 2017). This case shows the importance of measuring serum calcium in cattle that present signs compatible with neuromuscular syndromes, mainly in Mato Grosso do Sul, where other diseases such as botulism can cause similar conditions without macroscopic or histological changes.

1429The suspected *M. aeruginosa* poisoning, meanwhile, occurred in a batch of 780 steers, in which143035 died in less than 24 h after being introduced into a pasture where there was a dam. In the past, 601431cattle had previously died in the same way on the property. Three cattle were necropsied, one by the

1432 LAP-UFMS team and two by external veterinarians who sent samples for analysis. In all cases, the liver 1433 lesions were similar and were characterized by necrosis and marked hemorrhage in the centrilobular 1434 regions. The necropsied animal, in addition to the liver injury, had acute tubular nephrosis. 1435 Epidemiological characteristics such as the access of all affected cattle to the same water source, deaths 1436 concentrated over a short period of time, as well as the histological lesions identified are described in 1437 cases of poisoning by *M. aeruginosa*. However, the definitive diagnosis of this poisoning is made by 1438 detecting the microcystin toxin in the liver (Cullen & Stalker 2016), which was not performed in the 1439 present case.

1440 In relation to the suggestive diagnosis of alphavirus encephalomyelitis in horses, the necropsied 1441 animal had a negative result for rabies in direct immunofluorescence (IFD) and immunohistochemistry, 1442 presenting mononuclear and neutrophilic inflammation that were more pronounced in the gray matter 1443 of the cerebral cortex, thalamus, and cerebellum than in the spinal cord. Such histological lesions are 1444 compatible with those caused by alphaviruses, both in terms of inflammatory components and 1445 anatomical location (Luethy 2023). However, it was not possible to identify the etiological agent involved 1446 in the molecular tests carried out.

1447 Inconclusive diagnoses. Regarding the number of inconclusive diagnoses in necropsies carried 1448 out by the LAP-UFMS team (3/63), two were cases of perinatal death or the birth of a weak calf. This is a 1449 difficulty that occurs in most VDLs around the world, where the diagnosis rates are typically low and do 1450 not appear to have improved even with the development of new diagnostic techniques. Different causes 1451 can be involved in the occurrence of these losses, both infectious and non-infectious ones, and there is a 1452 need for a well-consolidated investigative triad involving the owner, the veterinarian, and the veterinary 1453 pathologist to improve such results (Mee 2020). This was, however, not possible since the cases received 1454 for necropsy or the field veterinarian did not characterize the loss in detail or sent the carcass in an 1455 advanced state of autolysis.

1456Regarding the total number of inconclusive diagnoses in this study, the number observed among1457necropsies carried out by the LAP-UFMS team or by veterinarians external to the LAP-UFMS (16.8%) is1458significantly lower than that observed in previous studies carried out in the same laboratory, either1459before the use of WhatsApp, in which 53.79% of necropsies resulted in inconclusive diagnoses (Pupin et1460al. 2019), or using the tool, in which 21.42% of necropsies resulted in inconclusive diagnoses (Lemos et1461al. 2023).

1462 Diseases that would not have been diagnosed without using WhatsApp. Among the diseases 1463 previously diagnosed at LAP-UFMS, but which, in this study, could only be diagnosed due to the 1464 possibility of the laboratory instructing the field veterinarian in real time via application regarding the 1465 best diagnostic procedure to be followed, four diseases deserve to be highlighted. Three of them refer to 1466 diseases that cause lesions to specific organs or sites, such as dictyocaulosis, eimeriosis, and secondary 1467 nutritional hyperparathyroidism in horses. In the first two diseases, the lesions and parasites are located 1468 in the lung, whereas eimeriosis also includes the ileum, cecum, and colon, and failure to evaluate these 1469 organs during necropsy with the collection and sending of materials for histological examination makes 1470 diagnosis impossible (Silva et al. 2005, Martins et al. 2020). In the case of dictyocaulosis, the diagnosis 1471 was made by visualizing the parasite specifically in the lung bronchi through photos. In the case of 1472 secondary nutritional hyperparathyroidism (fibrous osteodystrophy/"equine swollen face"), there were 1473 no significant histological lesions in the organ pieces sent, which did not include bones, and therefore, 1474 the characteristic lesions of fibrous osteodystrophy (Riet-Correa 2023) could not be observed. 1475 Consequently, the diagnosis was only possible by sending images and videos with clinical signs and 1476 necropsy findings (Figure 4). The other disease in this group is babesiosis, whose diagnosis was only 1477 possible due to the observation of suggestive clinical and pathological findings through photos and with 1478 a positive response to the implemented treatment.

1479 In the situations in which no necropsies were performed by the LAP team or materials were sent 1480 for histopathology, four situations were identified: (1) the photos sent were sufficient to make the 1481 diagnosis or suspicions; (2) the disease was diagnosed based on visual inspection during visits due to 1482 other causes; (3) therapeutic diagnosis - no necropsies were performed due to the positive response to 1483 treatment; and (4) it was impossible to collect material due to the lack of a veterinarian at the time of the 1484 deaths, the animals were found a long time after the deaths, or deaths in which the owner or veterinarian 1485 believes they already know the cause and do not require necropsy. It should be noted that these 1486 situations can be combined, for example, when there is a positive response to patient treatment and the 1487 patient subsequently dies, so that performing a necropsy is considered unnecessary.

1488Considering that the majority of disease surveys in Brazil are based on data from pathology1489laboratories, these diseases would not be recorded using necropsies alone (Lucena et al. 2010, Sprenger1490et al. 2015, Mello et al. 2017, Rondelli et al. 2017, Pupin et al. 2019, Molossi et al. 2021), and it is important

that LDVs develop systems capable of collecting and systematizing such information. In this group, the occurrences of anaplasmosis, haemonchosis, and multiple mineral deficiency in cattle and copper poisoning in sheep stand out, in which the suspected diagnosis was obtained based on the images sent and the diagnosis confirmed by ancillary laboratory tests and response to treatments. Two cases of hepatogenous photosensitization were observed in adult cows over 24 months old, both diagnosed based on visual inspection of the animals during visits to the property to attend to other health problems. The property's pasture was predominantly composed of *Brachiaria brizantha*, and no other possible causes of hepatogenic photosensitization were identified. Cases of hepatogenous photosensitization due to Brachiaria intoxication in cattle are frequent in Mato Grosso do Sul (Souza et al. 2010), but its occurrence is considered uncommon in adult cattle (Faccin et al. 2014). Observation in cases of other diseases shows that cases of hepatogenous photosensitization, especially when attributed to Brachiaria poisoning, are underreported. Dewlap edema, which develops seasonally in Mato Grosso do Sul, although it can affect a high number of cattle, does not cause productive losses or deaths. Therefore, this is also an underreported condition when we rely only on diagnoses from pathology laboratories. Another case is polioencephalomalacia, which, although it is a lesion and not a specific disease, is also an underreported problem because when its diagnosis is based on a positive response to treatment, it leads to no deaths and, consequently, no necropsies and notification.



 Figure 4. Secondary nutritional hyperparathyroidism in horses. Marked bilateral bulging of the face (fibrous osteodystrophy).

Under-reported occurrences. Among the complaints of field veterinarians who had sent images but did not send samples or animals for necropsy are abortions and neonatal diarrhea. In all these cases, several causes can be associated, and laboratory diagnosis is difficult. In the case of neonatal diarrhea, the number of deaths was lower than that of the animals that recovered, and establishing a precise diagnosis was not considered necessary by the owners. In this case, we also note the association between the behavior of ingesting soil (geophagy) by calves and the occurrence of diarrhea, which is not discussed in the literature. However, nutritional factors play an additional role in neonatal diarrhea, and inadequate nutrition can directly induce diarrhea or can further increase infectious diarrhea through osmosis or dysbiosis (Mol et al. 2022). Furthermore, this behavior is described in cases of mineral deficiencies such as sodium and iron deficiencies (Tokarnia 2010). Another important point regarding this problem is the conclusion by the field veterinarian that the cause of neonatal diarrhea is Escherichia *coli*, based only on the culture test performed on the feces. Such a diagnosis is mistaken since there are non-pathogenic strains of this bacterium in the ruminant microbiota and because it can also grow in culture; in addition, virulence factors can be identified in both diarrheal and normal feces (Tutija et al. 2022). Regarding abortions, three incidents were reported, all with images sent, but none with material sent (Figures 5-6). These data indicate that there was a loss of animals and of service time with the pregnant cow in these occurrences, showing economic losses. However, considering that abortion is a manifestation that can have several causes, including infectious agents, nutritional deficiencies, as well as genetic, toxic, and physical causes (Antoniassi et al. 2013, Mee. 2023), the establishment of a diagnostic approach that allows addressing all these different causes is fundamental for an assertive etiological diagnosis and the reduction of these losses. In one of the cases, although it was not possible to make a diagnosis regarding the etiology involved in the abortion, malformations such as arthrogryposis and increased skull volume (Figure 5) were identified. In these cases, some suspicions can be raised regarding the cause. Congenital malformations can occur as a result of hereditary genetic factors and/or environmental factors, which can be of infectious origin, when the female is infected by certain viruses, in cattle, mainly the bovine viral diarrhea virus (BVDV), or even nutritional and toxic in nature (Schild 2023).



Figures 5–6. Cases of abortions in cattle without etiological diagnosis. 5 – Bovine fetus with congenital malformations (arthrogryposis and increased cranial volume). 6 – Fetus in an advanced state of autolysis.

In contacts that did not result in diagnoses or suspicions by the LAP-UFMS due to the lack of information, apart from the notifier's complaint, some points in the field veterinarian's approach are noted. These include the high number of incidents in which the sender's suspicion or complaint was a neurological disease, including rabies. These data show the underreporting of suspected cases of this disease, which is a relevant fact since there is an official herbivore rabies control program. However, despite the existence of this program, the underreporting of rabies has been described previously (Mello et al. 2019). Another situation is the number of cases with suspected urea poisoning, many of which had a clinical picture and epidemiology compatible with this poisoning, but none of which had material sent for histopathology. This situation has previously been observed in the LAP-UFMS area of activity (Gimelli et al. 2023).

In numerous contacts where no material was sent, the suspicion was not of a specific disease, and generic terms were used to refer to clinical signs or injuries such as nervous signs, plant poisoning, mortality, coughing, skin lesions, pneumonia, polioencephalomalacia, and tremors. This behavior shows that the veterinarians themselves often do not understand the importance of clinical and epidemiological characterization when making a diagnosis. In this category, the erroneous use of the term "clostridiosis" was also observed, referring to a single etiological agent, whereas there is more than one species of *Clostridium* considered to be the causative agent of diseases in ruminants (Santos et al. 2019).

Diagnostic efficiency using WhatsApp. In addition to previously undiagnosed diseases and underreporting of diseases, this study shows an increase in the number of contacts via WhatsApp compared to a previous study that evaluated the same tool (Lemos et al. 2023). The previous study lasted 23 months, with 45 necropsies carried out by the LAP-UFMS team, 39 materials for histopathology, and 63 contacts without sending materials. In contrast, the present study lasted for 24 months, with 63 necropsies carried out by the LAP, 62 materials for histopathological analysis, and 86 contacts without sending materials. This increase in the number of contacts was not proportional to the increase in the number of veterinarians who had joined the application group, which was greater than 100%. Furthermore, the number of contacts that did not result in diagnoses due to samples or information not being sent was higher than that of animals sent for necropsy or samples for histopathology, individually, which demonstrates the need for improvements in communication via WhatsApp to increase the participation of veterinarians working in the diagnosis of diseases.

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CONCLUSION

1610 The use of WhatsApp facilitated communication and promoted different forms of contact between 1611 field veterinarians and the veterinary diagnostic laboratory, which increased the number of conclusive 1612 diagnoses and allowed the identification of diseases not previously diagnosed in Mato Grosso do Sul, 1613 diseases previously diagnosed but which occurred in different production systems, and underreported 1614 diseases. It also allows the continuous monitoring of outbreaks, which allowed underestimating the 1615 number of disease occurrences. Furthermore, it was possible to identify the use of mistaken diagnostic 1616 approaches and concepts by field veterinarians as well as the occurrence of health problems with challenging diagnoses, such as abortions, perinatal deaths, and deaths by neonatal diarrhea. 1617

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CAPÍTULO 5 – Atividades complementares
4.1 Capítulo do Boletim anual do Laboratório de Anatomia Patológica da Universidade Federal de Mato Grosso do Sul
A utilização do WhatsApp® para
identificação de doenças não diagnosticadas
previamente e subnotificadas de ruminantes
e equinos em Mato Grosso do Sul
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Figura1. Capítulo do Boletim anual do Laboratório de Anatomia Patológica da Universidade Federal de Mato Grosso do Sul
Neste capítulo do Boletim Anual do Laboratório de Anatomia Patológica da
Universidade Federal de Mato Grosso do Sul, foi abordado sobre como o whatsapp
foi uma ferramenta que ajudou a melhorar a eficiência dos diagnósticos,
identificando doenças que antes não eram diagnosticadas. A equipe reuniu neste
boletim todos os diagnósticos realizados em animais de produção durante o ano de
2023, buscando contribuir de maneira substancial para a compreensão e para o
desenvolvimento de estratégias de tratamento mais eficazes.
4.2 Notas técnicas
O objetivo principal é fornecer informações técnicas atualizadas e relevantes
aos veterinários que atuam em campo no estado de Mato Grosso do Sul, Brasil, e
em países vizinhos como Paraguai e Bolívia. Essas notas técnicas são
desenvolvidas diretamente a partir das demandas identificadas pelos profissionais
que lidam diariamente com diagnósticos e controle de doenças em animais de
produção.

Em termos práticos, as notas técnicas podem oferecer suporte rápido e direto aos profissionais de campo, capacitando-os a tomar decisões mais informadas e assertivas no manejo de saúde animal. Ao melhorar a eficiência dos laboratórios de diagnóstico e a comunicação entre os veterinários, esperamos contribuir significativamente para a sanidade animal e para a segurança alimentar na região.



CONSIDERAÇÕES FINAIS

O uso do Whatsapp® como ferramenta ajudou a melhorar significativamente 1837 a eficiência dos diagnósticos e a comunicação entre o veterinário de campo e o 1838 LAP-UFMS, proporcionando um aumento nos diagnósticos conclusivos em relação 1839 1840 ao ano anterior, possibilitou a identificação de doenças anteriormente não diagnosticadas na região, permitiu reconhecer doenças em diferentes sistemas de 1841 produção animal, facilitou o monitoramento de surtos e a rápida comunicação de 1842 casos, evitando subnotificações, identificou práticas 1843 novos diagnósticas equivocadas possibilitando correções e melhorias nos métodos utilizados e auxiliou 1844 na investigação de problemas sanitários desafiadores como abortos, mortes 1845 1846 perinatais e mortes por diarreia. Os materiais obtidos através da metodologia implementada são uma importante fonte para o laboratório e podem ser 1847 transformados em publicações como artigos científicos, notas técnicas e boletins, 1848 disseminando os resultados e contribuindo com o avanço do conhecimento da 1849 1850 comunidade científica e técnica.

- 1851
- 1852 1853

INOVACÃO

ECONÔMICO, SOCIAL, TECNOLÓGICO IMPACTO E/OU

As estratégias propostas neste estudo foram eficazes em melhorar 1854 а eficiência e a comunicação do LAP com os veterinários e vice-versa. O impacto 1855 1856 econômico gerado foi a agilidade nos processos. Isso reduziu custos operacionais, aumentou a produtividade e diminuiu o tempo de resposta aos requisitantes. O 1857 impacto social gerado foi a melhor comunicação e satisfação do usuário do serviço 1858 do laboratório, garantindo um suporte mais personalizado e eficiente. Os materiais 1859 didáticos produzidos servem para educação continuada para os veterinários 1860 1861 parceiros, isso ajuda a melhorar a qualidade do serviço prestado e aumentar a conscientização sobre sanidade animal nesse meio profissional. Quanto ao impacto 1862 tecnológico, o uso do Whatsapp® permitiu o agendamento de necropsias, 1863 comunicação de resultados e acesso facilitado à informações, isso facilitou o fluxo 1864 de trabalho. A inovação foi usar uma ferramenta de mídia social para diagnóstico 1865 de forma padronizada e sistematizada. A implementação do Whatsapp® funcionou 1866 como um sistema robusto de resposta aos veterinários, gerando dados passíveis 1867

1868 de serem utilizados para identificar áreas de melhoria e oportunidades para 1869 inovação.