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Acute necrotic hepatotoxicity caused by *Lantana* camara L. ingestion in dairy cattle¹

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ABSTRACT.- Silva E.M.S., Rosa R.B., Molossi F.A., Ribeiro P.R., Almeida K.G.S., Pinheiro G.F., Dalto A.G.C., Panziera W. & Pavarini S.P. 2021. **Acute necrotic hepatotoxicity caused by** *Lantana camara* L. ingestion in dairy cattle. *Pesquisa Veterinária Brasileira* 41:e06893, 2021. Setor de Patologia Veterinária, Departamento de Patologia Clínica Veterinária, Faculdade de Veterinária, Universidade Federal do Rio Grande do Sul, Av. Bento Gonçalves 9090, prédio 42505, Porto Alegre, RS, 91540-000, Brazil. E-mail: saulo.pavarini@ufrgs.br

This study describes an outbreak of acute necrotic hepatopathy associated with spontaneous poisoning by *Lantana camara* L. in dairy cattle. A herd of 15 cows and heifers was introduced into a native pasture with limited food supply, and, sixteen days later, eight animals had spontaneous nasal hemorrhage, fever, lethargy, jaundice, and dry, dark stools with mucus and blood. The clinical course varied from two to five days. In the pasture where the cattle were kept, abundant adult specimens of *L. camara* L. with evident signs of consumption were observed. In total, seven cattle died and necropsy was performed in three of them. All animals had moderate jaundice, hemorrhage in the subcutaneous tissue and on the surface of different organs. The liver was slightly enlarged, with orange discoloration and enhanced lobular pattern. Histologically, multifocal areas of coagulative necrosis of hepatocytes in the centrilobular area, occasionally extending to the midzonal area, were observed, as well as marked hepatocellular degeneration and prominent cholestasis. The current study suggests that *L. camara* L. poisoning should be considered a differential diagnosis of acute and necrotic hepatotoxicity in cattle, despite the absence of photosensitization.

INDEX TERMS: Necrotic hepatotoxicity, acute intoxication, liver, necrosis, *Lantana camara* L., toxic plants, dairy cattle, plant poisoning.

RESUMO: - [Hepatotoxicidade necrótica aguda por ingestão de *Lantana camara* L. em bovinos leiteiros.] Esse estudo descreve um surto de hepatopatia necrótica aguda associada a intoxicação espontânea por *Lantana camara* L. em bovinos leiteiros. Um lote de 15 vacas e novilhas foram introduzidas em um piquete com campo nativo, com escassa oferta de alimento. Após dezesseis dias, oito animais manifestaram epistaxe, febre, apatia, icterícia, fezes ressecadas e escuras com muco e sangue. A evolução do quadro clínico variou de

dois a cinco dias. No piquete em que os bovinos estavam alojados havia grande quantidade de *L. camara* L. com sinais evidentes de consumo. No total, sete bovinos morreram, e destes, o exame *post mortem* foi realizado em três. Os bovinos exibiam moderada icterícia, hemorragias no tecido subcutâneo e na superfície de diferentes órgãos. O fígado estava discretamente aumentado, com coloração alaranjada e padrão lobular evidente. As lesões histológicas consistiam em acentuada necrose de coagulação de hepatócitos em região centrolobular, por vezes se estendendo a região mediozonal, além de acentuada degeneração dos hepatócitos e evidente Colestase. O presente trabalho alerta para que intoxicação por *L. camara* L. seja levada em consideração nos diagnósticos diferenciais de hepatotoxicidade necrótica aguda em bovinos, mesmo sem indícios de fotossensibilização.

TERMOS DE INDEXAÇÃO: Hepatotoxicidade necrótica, intoxicação aguda, fígado, necrose, *Lantana camara* L., plantas tóxicas, bovinos leiteiros.

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INTRODUCTION

Lantana species are cosmopolitan, ornamental, exotic, and invasive plants; they are widely distributed in Brazil, from Amazon to Rio Grande do Sul (Tokarnia et al. 2012). There are few reports in Brazil of spontaneous poisoning by ingestion of *Lantana* sp. in cattle (Riet-Correa et al. 1984, Tokarnia et al. 1984, 1999, Rissi et al. 2007, Lucena et al. 2010) and buffalo (Bastianetto et al. 2005). Nonetheless, experimental poisoning has been reproduced in cattle (Riet-Correa et al. 1984, Tokarnia et al. 1984, 1999), sheep (Brito & Tokarnia 1995), and rabbit (Brito 1995). In countries such as Australia and South Africa, Lantana camara is considered one of the most important toxic plants for cattle (Kellerman et al. 2005, Tokarnia et al. 2012). Primary conditions for natural poisoning include shortage of forage and relocation of animals to pastures with high L. camara density (Riet-Correa et al. 1984). L. camara, Lantana tiliaefolia, and Lantana glutinosa are the main species involved in cases of poisoning in animals in Brazil (Tokarnia et al. 2012), and the toxicity is caused by toxic triterpenes lantadene A and lantadene B, which damage the hepatocytes and bile canaliculi (Low 1948, Tokarnia et al. 2012).

The classic clinical signs of *L. camara* poisoning in cattle are severe jaundice and secondary photosensitization, with histological findings of hepatic fatty degeneration and marked cholestasis (Tokarnia et al. 2012). However, there are few reports of hepatic necrosis due to acute poisoning by *L. camara* in ruminants (Fourie et al. 1987, Ide & Tutt 1998). In the cases reported in these studies, the animals had jaundice, anorexia, and ruminal stasis, and the clinical signs developed within 24 to 48 hours; however, there were no signs of photosensitization because of early death from severe liver damage (Tokarnia et al. 2012). Therefore, the aim of this study was to describe the epidemiological, clinical, and pathological aspects of an outbreak of acute necrotic hepatotoxicity by *L. camara* L. in dairy cattle in Rio Grande do Sul.

MATERIALS AND METHODS

Visits to the farm were performed during the disease outbreak, wherein epidemiological data collection and observation of clinical signs were carried out, as well as necropsy of three cows. Samples from multiple organs (liver, spleen, central nervous system, lung, heart, skeletal muscle, kidney, intestines and pre-stomachs) were collected, fixed in 10% formalin, and processed for histological evaluation with hematoxylin and eosin (HE) staining. In addition, leaf and flower samples from *Lantana* spp. specimens were collected and sent to the "Departamento de Botânica", "Herbário do Instituto de Ciências Naturais" of the "Universidade Federal do Rio Grande do Sul" (UFRGS) for morphological analysis and botanical identification. These plant samples were registered under the number 202187.

RESULTS

The cases of acute and necrotic hepatotoxicity were observed on an 8-hectare rural property in the city of Viamão, Rio Grande do Sul (30°4′51″ S, 51°1′22″ W) in July 2020. A batch of 15 Holstein cattle, all females (heifers and dry cows) aged from 1.5 to 8 years old, was introduced to a 1.2-hectare area with scarce native pasture and without mineral and concentrate supplementation. The pasture had forest and shrub vegetation close to the fence outlining the area. During the visits, several specimens of *Lantana* spp., with evident consumption were

observed (Fig.1). The plant species was identified as *Lantana camara* L. based on morphological analysis. Moreover, other plants (*Cestrum intermedium*, *Cestrum parqui*, *Xanthium* spp. and *Dodonaea viscosa*) and larvae (*Perreyia flavipes*), which may cause acute liver necrosis in the region, were not observed on the property.

Eight of the fifteen cattle (morbidity: 53%) had spontaneous nasal hemorrhage, fever over 40°C (reference value: 38.5-39.5°C) (Constable et al. 2017), lethargy, jaundice, and dry, dark stools with mucus and blood; one of the cows had a an abortion. The animals started with clinical signs after sixteen days in the pasture with specimens of *Lantana* spp., and the clinical course varied from 2 to 5 days after relocation of the animals. In total, seven cows died (mortality: 46% and lethality: 87%); however, after 3 months of the outbreak, affected and non-affected cattle had signs of photosensitization.

Hepatic and extrahepatic changes observed in the three necropsied cows (Cattle 1, 2, and 3) (Table 1) were similar and included jaundice of the omentum and the serosal surface of multiple organs; mild subcutaneous edema, mainly in the ventral region; and moderate ascites with varying severity of hemorrhage (petechiae and suffusions) affecting the omentum, abomasal serosa, pre-stomachs, small and large intestines, gall bladder (Fig.2), and heart (pericardium and endocardium). Hemorrhage in the splenic capsule was also observed in Cattle 3. The liver in Cattle 3 was mildly enlarged with enhanced lobular pattern and the gallbladder had multifocal to coalescent serosal hemorrhages (Fig.3). On the cut surface, all evaluated



Fig. 1. Acute necrotic hepatotoxicity by *Lantana camara* L. in dairy cattle. Shrub vegetation of *L. camara* L. with evidence of consumption. Inset: *L. camara* leaves and inflorescences collected in the local of the outbreak.

Table 1. Necropsy findings of acute liver poisoning by Lantana camara L. in cattle

Cattle	Jaundice	Enhanced lobular pattern	Liver discoloration		
1	++	+++	Diffusely orange		
2	++	+++	Diffusely orange		
3	+++	+++	Yellowish areas interspersed with red		

⁺ Mild, ++ moderate, +++ severe.

livers were friable and had two distinct macroscopic patterns: slightly irregular, with predominant orange discoloration in Cattle 1 and 2 (Fig.4), and yellowish areas interspersed with dark and red foci in Cattle 3 (Fig.5).

Histologically, classic findings of acute hepatotoxicity were observed in all cases (Table 2). Degenerative lesions were prominent in the periportal hepatocytes and were characterized

by marked intracytoplasmic micro- and macro-vacuolar degeneration, which was often associated with moderate neutrophilic inflammatory infiltrate (Fig.6). Marked cholestasis in the bile canaliculi and ducts (Fig.7) and occasional small bile accumulations within the hepatic parenchyma were observed, as well as mild periportal fibrosis and bile duct proliferation (Fig.8). The liver of Cattle 3 had marked hepatocellular coagulative

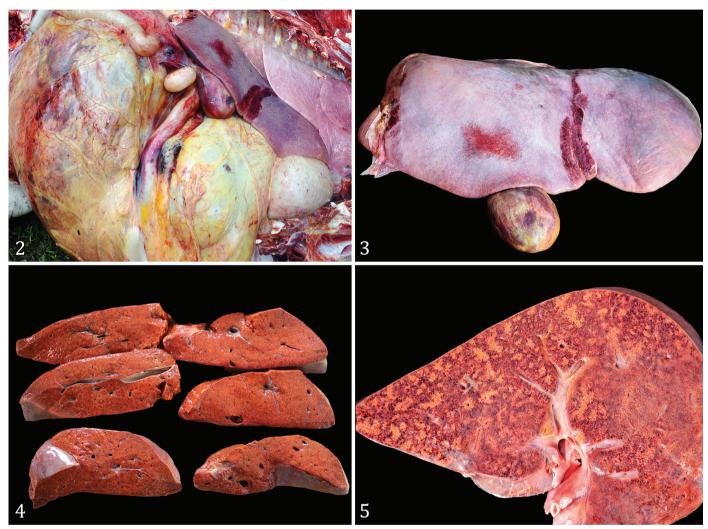


Fig.2-5. Macroscopic findings of acute *Lantana camara* L. poisoning in dairy cattle. (2) Diffuse jaundice of omentum, serosal surface of multiple organs, multifocal hemorrhages in the omentum, serosa of the abomasum, liver and gallbladder (Cattle 3). (3) Mild hepatomegaly, enhanced lobular pattern and multifocal areas of hemorrhage on the hepatic capsular surface, in addition to multifocal to coalescent serosal hemorrhage in the gallbladder (Cattle 3). (4) Liver irregular cut surface (Cattle 1), orange discoloration and enhanced lobular pattern. (5) Liver cut surface (Cattle 3), heterogeneous in appearance, with yellowish areas interspersed with dark and light red foci.

Table 2. Histopathological findings of acute necrotic hepatotoxicity by Lantana camara L. in dairy cattle

Cattle	Coagulative necrosis	Cholestasis	Vacuolar degeneration	Bile duct proliferation	Hemorrhage	Neutrophilic inflammatory infiltrate	Periportal fibrosis
1	Random ++	++	+++	+	++	+	+
2	Centrilobular +++	+++	+++	+	++	++	+
3	Centrilobular to midzonal +++	+++	++	+	+++	++	+

⁺ Mild, ++ moderate, +++ severe.

necrosis and hemorrhage, which were predominantly in the centrilobular area, frequently extending to the midzonal area, and bridging across the hepatic lobule (Fig.8 and 9).

DISCUSSION AND CONCLUSION

In the current study, the diagnosis of acute hepatotoxicity in dairy cattle caused by ingestion of *Lantana camara* L. was based on epidemiological data, clinical signs, macroscopic and histologic evaluation of affected cattle, in addition to the detection of large quantities of plant specimens with evidence of consumption. Although outbreaks of acute necrotic hepatopathy caused by *Lantana* spp. poisoning are severe, it is uncommon in Brazil, because this plant is unpalatable and clinical cases only occur with low pasture availability, as during winter in Southern Brazil, or with relocation of animals to pastures with high density of the plant (Brito & Tokarnia 1995, Tokarnia et al. 1999, Brito et al. 2004, Lucena et al. 2010). All of these factors indicate a strong association with

the outbreak of acute necrotic hepatopathy by *L. camara* L. poisoning in the cattle described in this study.

Lantana species are classified as hepatotoxic plants that cause hepatogenous photosensitization (type III) and jaundice in livestock (Kellerman et al. 2005, Tokarnia et al. 2012). However, in the present study, affected animals had acute liver damage without manifestations of photodermatitis, which represents an unusual clinical presentation of *Lantana* poisoning that is rarely reported in cattle (Fourie et al. 1987). Some authors described that development of acute liver changes may be correlated with the ingested dose and the time needed for evolution of clinical signs (Fourie et al. 1987, Ide & Tutt 1998, Tokarnia et al. 2012). Additionally, some studies reported that the occurrence and intensity of photodermatitis can be variable in the different species of animals affected by Lantana spp. (Sharma et al. 2007, Gupta et al. 2019). The absence of photosensitization in cattle in this study is similar to previous cases in cattle (Seawright & Allen 1972) and goats

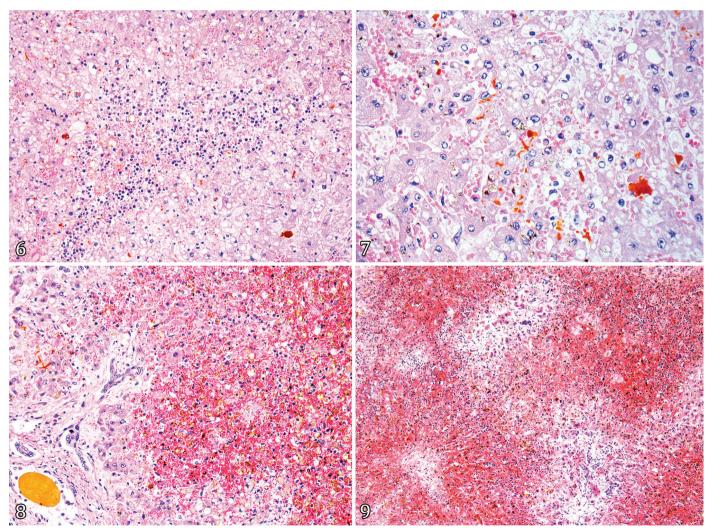


Fig.6-9. Hepatic histological findings of acute *Lantana camara* L. poisoning in dairy cattle. (6) Centrilobular necrosis with hemorrhage and neutrophilic inflammatory infiltrate. Adjacent to this area, there is marked hepatocellular cytoplasmic vacuolation and prominent cholestasis (Cattle 2). HE, obj.20x. (7) Evidence of cholestasis in canaliculi and bile ducts and severe hepatocellular cytoplasmic vacuolation (Cattle 2). HE, obj.40x. (8) Bridging coagulative necrosis and marked hemorrhage in the centrilobular area, extending to the midzonal area, forming bridges. There are also mild periportal fibrosis and bile duct proliferation (Cattle 3). HE, obj.20x. (9) Necrosis and hemorrhage of the hepatic parenchyma with hepatocellular vacuolization and prominent cholestasis (Cattle 3). HE, obj.10x.

(Obwolo et al. 1991, Ide & Tutt 1998), and may be related to the acute onset of the disease, in which accumulation of phylloerythrin in plasma and exposure to the sunlight were insufficient to develop skin lesions. In severe cases, animals can die before showing clear signs of photosensitization, as in the current study (Ide & Tutt 1998, Sharma et al. 2007). Studies revealed that the lethal dose may vary from 5g/kg to 40g/kg (Tokarnia et al. 1999, Brito et al. 2004), but in the present investigation it was not possible to determine the dose that caused an acute death, mainly because the animals were grazing and it may have had dominant cattle. However, there were considerable amounts of consumed plant, and food scarcity was also observed. Another factor that may have influenced the unusual presentation of the disease is the range of toxicity of *Lantana* species, which may vary according to each region (Brito et al. 2004, Tokarnia et al. 2012). Furthermore, abortion has been reported in an animal poisoned by the plant, but a correlation could not be established (Riet-Correa et al. 1984, Tokarnia et al. 1984).

The cause of death in cattle in this study was attributed to acute liver failure due to *L. camara* L. poisoning. Although gross and histologic lesions of hepatocellular necrosis in cases of *L. camara* poisoning are uncommon in cattle, the lesions are similar to those observed in another study described in bovine species (Fourie et al. 1987). The orange to yellow discoloration of the liver observed in the affected cattle pointed to poisoning by *Lantana* spp. Subcutaneous edema was discarded as evidence of photosensitization because it wasn't located in the depigmented areas of the skin and no erythema, flaking and necrosis were observed (Tokarnia et al. 1984). In addition, moderate ascites and subcutaneous edema are believed to have occurred because of severe liver injury with impaired hepatic function (Mosier 2017).

Histologically, in addition to necrotic liver changes and hemorrhage, degenerative lesions and cholestasis were prominent and have been reported in L. camara poisoning in cattle (Riet-Correa et al. 1984, Tokarnia et al. 1984, 1999). Although other toxic plants have similar histopathological findings of hepatic liver necrosis and vacuolar degeneration, prominent cholestasis is not observed, which is a characteristic lesion of *Lantana* spp. poisoning (Tokarnia et al. 2012). Extrahepatic changes supported the diagnosis of jaundice and hemorrhage in several organs. Necrosis of the renal tubular epithelium is also described (Tokarnia et al. 2012); however, no renal lesions were observed in the affected cattle described in this study. It is known that in Lantana camara poisoning, the lesion of hepatocellular necrosis and degeneration starts in hepatocytes in the periportal region and extend to the centrilobular region, mainly as a result of the interaction of toxins with hepatocyte receptors, which causes bile regurgitation and cholestasis. Subsequently, a chronic lesion of hyperplasia, bile duct proliferation and fibrosis in the periportal area develops, which may justify the histologic findings of the present study (Sharma et al. 1981, 1988, 2007). The molecular events in hepatocytes that trigger the toxic effect are not yet known, but electron microscopy analysis revealed hepatocytes with dispersed, fragmented and vacuolized endoplasmic reticulum (Seawright 1965).

Lantana camara poisoning in cattle must be differentiated from other conditions that cause acute toxic liver disease in the region of outbreak, such as Cestrum poisoning caused

by C. intermedium (Furlan et al. 2008, Bandarra et al. 2009, Wouters et al. 2013) or *C. corymbosum* var. hirsutum (Gava et al. 1991), and C. parqui (Riet-Correa et al. 1986); Xanthium cavallinesii poisoning (Driemeier et al. 1999); Dodonaea viscosa poisoning (Colodel et al. 2003); and *Perrevia flavipes* larvae poisoning (Soares et al. 2008, Raymundo et al. 2009, Tessele et al. 2012). Although subacute poisoning by Senecio spp. is uncommon (Tokarnia & Döbereiner 1984, Panziera et al. 2017), it should also be considered for differential diagnosis. These causes were discarded in the current cases because these plant species and larvae were not present in the area accessible to the cattle. Additionally, morphological changes, including hepatocellular degeneration, cholestasis, orange discolored liver, and degenerative lesions are characteristic of Lantana spp. poisoning in cattle (Riet-Correa et al. 1984, Tokarnia et al. 1984, 1999). Although copper poisoning in cattle may have similar histologic lesions in the liver, such as centrilobular necrosis, degeneration and variable cholestasis, macroscopically, the liver has enhanced lobular pattern with non-orange discoloration, different from that observed in Lantana spp. poisoning. Also, copper poisoning in cattle is associated with copper supplementation, which was not performed in the animals of this study, and have lesions related to intravascular hemolysis, a feature not present in the current cases (Martins et al. 2020). Thus, L. camara L. poisoning must be considered as a differential diagnosis in cases of acute necrotic hepatopathy in cattle, despite the absence of photosensitization.

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 $\textbf{Conflict of interest statement.-} \ The \ authors \ have \ no \ competing \ interests.$

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