

Conditioned food aversion to control *Palicourea aeneofusca* poisoning

Aversão alimentar condicionada para o controle da intoxicação por *Palicourea aeneofusca*

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– NOTE –

ABSTRACT

Palicourea aeneofusca contains sodium monofluoroacetate, which causes sudden death in ruminants when administered at doses of approximately 0.6g kg⁻¹ of body weight (g kg⁻¹). In this experiment two groups of 6 goats were used to determine the possibility to induce conditioned food aversion to *P. aeneofusca*. In group 1, 0.35g kg⁻¹ of green leaves of the plant were given to six goats on days 1, 5, 10, 20, 30, 60, and 90 of the experiment. On the first day, all of the goats ingested the full amount of the plant and were treated immediately with 175mg kg⁻¹ of lithium chloride (LiCl) through a ruminal tube. On day 5, only two goats ingested the plant, and they were treated with the same dose of LiCl. On days 10, 20, 30, 60, and 90, none of the goats ingested the plant. For another group of 6 goats, the leaves were given on days 1, 10, 20, 30, 60, and 90. All of the goats ingested the leaves on day 1 and received 1mL kg⁻¹ body weight of water through a ruminal tube. All of these goats ingested the plant on days 10, 20, 30, 60, and 90. These results demonstrate that it is possible to induce conditioned food aversion to *P. aeneofusca* that persists for at least 90 days. Further experiments should be performed to determine the duration of the aversion and to induce aversion to other *Palicourea* species, particularly *P. marcgravii*, which is the most important toxic plant in Brazil.

Key words: poisonous plants, *Palicourea* spp., sodium monofluoroacetate, conditioned food aversion.

RESUMO

Palicourea aeneofusca, que contém monofluoroacetato de sódio, causa morte súbita em ruminantes quando é administrada a doses de aproximadamente 0,6g kg⁻¹ de peso vivo (g kg⁻¹). Neste experimento, foram utilizados dois grupos de 6 caprinos para determinar a possibilidade de induzir aversão alimentar condicionada à ingestão de *P. aeneofusca*. Para induzir aversão alimentar condicionada no grupo 1, 0.35g kg⁻¹ de folhas

verdes da planta foram administradas a seis caprinos nos dias 1, 5, 10, 20, 30, 60, e 90 do experimento. No primeiro dia, todos os caprinos ingeriram toda a planta oferecida e foram tratados imediatamente com 175mg kg⁻¹ de carbonato de lítio (LiCl) através de sonda ruminal. No dia 5, somente dois caprinos ingeriram a planta e foram tratados com a mesma dose de LiCl. Nos dias 10, 20, 30, 60, e 90 nenhum caprino ingeriu a planta. Seis caprinos do grupo controle receberam 0.35g kg⁻¹ de folhas nos dias 1, 10, 20, 30, 60, e 90. Todos os caprinos ingeriram as folhas no dia 1 e foram tratados com 1mL kg⁻¹ pv de água mediante sonda ruminal. Todos os caprinos deste grupo tornaram a ingerir a planta nos dias 10, 20, 30, 60 e 90. Esses resultados demonstram que é possível induzir aversão alimentar condicionada à *P. aeneofusca*, que persiste por pelo menos 90 dias. Próximos experimentos deverão ser realizados para determinar a duração da aversão e para induzir aversão contra outras espécies de *Palicourea*, particularmente *P. marcgravii*, que é a planta tóxica mais importante do Brasil.

Palavras-chave: plantas tóxicas, *Palicourea* spp., monofluoroacetato de sódio, aversão alimentar condicionada.

Palicourea aeneofusca (Müll. Arg.) Standl., known as *cafezinho* or *erva-do-rato*, causes sudden death associated with exercise in cattle in the coastal regions of Pernambuco, Bahia, Alagoas, and Paraíba (VASCONCELOS et al., 2008). Experimentally administered *P. aeneofusca* is toxic to cattle, rabbits and goats (PASSOS, 1983). Its toxicity is caused by sodium monofluoroacetate (MFA), which inhibits aconitate hydratase, one of the tricarboxylic acid cycle enzyme (LEE et al., 2012). Among the plants that contain MFA, those from the

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Rubiaceae family, including *Palicourea marcgravii*, *Palicourea grandiflora*, *Palicourea juruana*, and *P. aeneofusca*, are the main toxic plants in Brazil that affect livestock. *P. marcgravii* occurs throughout most of Brazil, except in states of the southern region, the State of Mato Grosso do Sul, and the semiarid region of Northeastern Brazil, and is the most important toxic plant with Brazil (TOKARNIA et al., 2012). The lethal dose of fresh *P. aeneofusca* leaves for goats and cattle (TOKARNIA et al., 1983) is 0.6 and 0.75g kg⁻¹ of body weight (g kg⁻¹), respectively. The toxicity of *P. marcgravii* was found to be similar, i.e., 0.5 to 0.6g kg⁻¹, in different experiments using cattle, goats, and sheep. In addition to its wide geographical distribution and high toxicity, intoxication by *P. marcgravii* is also due to its high palatability, which makes animals consume it even when it is sparse. *P. marcgravii* has a cumulative effect when administered daily, but after consuming the plant, the MFA that it contains seems to be detoxified in 2-5 days (TOKARNIA et al., 2012). The clinical signs shown by animals that have been poisoned by *Palicourea* spp. are tachycardia, jugular engorgement with a venous pulse, tachypnea, a staggering gait, falls, and recumbence, followed by death. No significant macroscopic lesions have been reported. Histologically, the only significant lesions are vacuolar hydropic degeneration and pyknosis of the epithelial cells in the distal convoluted tubes of the kidneys (PASSOS, 1983).

Conditioned food aversion is a technique used with ruminants and horses to prevent their ingestion of poisonous plants. To accomplish this, the emetic substance lithium chloride (LiCl) is administered through a ruminal fistula or gastric tube immediately after the animal consumes the plant; the animal associates the illness caused by LiCl with the taste of the food and subsequently avoids that taste (RALPHS et al., 2001). In Brazil, conditioned food aversion has been used experimentally to prevent the ingestion of *Leucaena leucocephala* (GÓRNIK et al., 2008), *Amorimia* (*Mascagnia*) *rigida* (BARBOSA et al., 2008) and *Ipomoea carnea* subsp. *fistulosa* (PIMENTEL et al., 2012). Field experiments have demonstrated the usefulness of this technique on commercial farms to prevent the intake of *Turbina cordata* and *I. carnea* (PIMENTEL et al., 2012; OLIVEIRA et al., 2013). These low-toxicity plants are offered in repeated daily doses to induce aversion, and LiCl is administered each time the animals ingest the plants (PIMENTEL et al., 2012). There are no studies demonstrating the possibility of inducing aversion to plants that are highly toxic and have a cumulative effect, such as *Palicourea* spp.

This study aimed to determine whether it is possible to induce aversion to *P. aeneofusca* by administering very low doses of the plant (approximately 50% of the toxic dose) at intermittent periods, allowing MFA to be detoxified in the intervals between each administration.

The experiment was conducted in the Laboratory of Goat Science, at the Federal University of Paraíba (UFPB) located in the city Bananeiras, Paraíba. The plants were collected at the Center for Agricultural Sciences of the UFPB in the city of Areia, Paraíba (S07°04'02" W37°16'51" and at an altitude of 567 m above sea level). The plants were collected the day before use, packed in nylon bags, and stored under refrigeration until administration. The MFA concentration in samples of *P. aeneofusca* collected in the same location was 0.09±0.05% (LEE et al., 2012). Twelve 12- to 18-month-old Saanen goats were used in the experiment. The goats were divided into two groups of six animals. The goats in Group 1, weighting 18, 19, 25, 23, 18, and 25kg, were averted with LiCl after ingesting *P. aeneofusca*, and the goats in Group 2, weighting 18, 24, 18, 31, 17, and 24kg, received water after first consuming the plant. Each goat was placed in an individual pen. During the experiment, the animals received *Pennisetum purpureum ad libitum* and 300g day⁻¹ of a concentrate consisting of 40% ground corn, 20% soybean meal, 20% wheat bran and 20% cottonseed meal.

Before aversion treatment, the animals were fasted for 12 hours. They were then each weighed, and green leaves of *P. aeneofusca* at a dose of 0.35g kg⁻¹ were administered during 10 minutes. The goats in Group 1 that ingested the plant were treated within 10 minutes with 175mg kg⁻¹ of LiCl administered through a gastric tube. The LiCl solution contained 175 mg of LiCl per 1 mL of water. The leaves were offered on days 1, 5, 10, 20, 30, 60, and 90. The goats in Group 2 (control) were fed 0.35g kg⁻¹ of leaves of *P. aeneofusca* on days 1, 10, 20, 30, 60, and 90. On the first day on which the plant was offered, plant ingestion was followed by the administration of 1mL kg⁻¹ of water through a gastric tube. After the administration of LiCl or water, the animals were observed for the presence of any clinical signs and the heart and respiratory rates were evaluated.

On the first day of the experiment, all of the goats in Group 1 spontaneously ingested all of the *P. aeneofusca* leaves offered (0.35g kg⁻¹) and were immediately treated with LiCl. On the fifth day, only two animals ingested the leaves and were treated with LiCl. On days 10, 20, 30, 60, and 90, none of the

animals in this group ate the plant. All of the goats in Group 2 ingested all the leaves on day 1 and received 1 mL kg⁻¹ of water through a gastric tube. On days 10, 20, 30, 60, and 90, all of the animals ingested all of the leaves offered. Clinical signs were not observed in any goat from both groups during the experiment.

These results indicate that it is possible to efficiently induce conditioned food aversion to *P. aeneofusca*. Prior to this experiment, the main limitation to the use of conditioned food aversion for the prophylaxis of poisoning by this plant was its high toxicity, which limited the amount of the plant that could be offered before the administration of LiCl. In this experiment, the ingestion of only 0.35g kg⁻¹ of the plant followed by the administration of LiCl was extremely effective in inducing aversion; a single administration to four of the six goats in Group 1 and two administrations to the remaining two animals induced aversion. The other important result was that the aversion persisted during the 90 days of experiment, despite the extreme palatability of the plant. In a study on inducing aversion to *Leucaena leucocephala* in goats, which is highly palatable, the amount of the plant the treated goats consumed was considerably reduced, but they continued consuming small amounts of it (GÓRNIK et al., 2008).

Field experiments should be conducted to test the efficacy of applying conditioned aversion to *P. aeneofusca* and other species of *Palicourea*, particularly *P. marcgravii*, which is the major toxic plant in Brazil (TOKARNIA et al., 2012). Obviously, it is essential to establish whether the dose of 0.35g kg⁻¹ is not toxic to the animals before starting the aversion process in a herd. In this experiment, aversion lasted at least 90 days. Studies with other plants demonstrated that aversion lasted up to three years (RALPHS & PROVENZA, 1999). Studies conducted in Northern and Northeastern Brazil demonstrated that it is possible to induce aversion to *Turbina cordata* and *Ipomoea carnea* that persists for periods of up to two years (OLIVEIRA et al., 2013).

In conclusion, it is possible to use conditioned food aversion to prevent the consumption of and intoxication by *P. aeneofusca*, and most likely, this technique can be applied to prevent ruminants from consuming *P. marcgravii*. Field experiments should be conducted to confirm these possibilities.

ETHICS COMMITTEE

The experiment was approved by the ethical committee on animal experimentation on the UFCG, process CEP 69-2013.

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