

## Leukogram and Platelet Values of Experimental Ovines Tested With Buds of Bracken Fern (*Pteridium Aquilinum*)

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#### Abstract

Bracken fern (Pteridium aquilinum) is an important toxic plant that causes three different forms of diseases, two chronic forms that cause digestive system and bladder tumors and an acute form that causes hemorrhagic diathesis. Cattle develop the acute form of the disease when they ingest doses above 10 g/kg a day, and hematology showed leukopenia and thrombocytopenia. There is a lack of information in the literature on the effect of the acute form of the disease caused by this plant in ovines. Here, we report the clinical and hematology parameters of two ovines that received buds of bracken fern in daily doses of 8.5 g/kg and 11.8 g/kg for 16 days and were monitored for 4 weeks after ceasing intake. During the experimental period, there was no change in clinical or hematological parameters. The parameters of the leukocyte count and platelets were normal, suggesting that the bracken fern is not toxic to the species at the doses tested, during the length of the intake period, and at the time of year the plant was harvested.

Keywords: Bracken fern; Ovine; Leukocytes; Platelets

#### Introduction

Bracken fern (*Pteridium aquilinum*) is an important toxic plant, found primarily in the southern region of Brazil [1-4]. The toxicity of the plant in cattle is already documented, and ingestion can cause chronic and acute intoxication: two chronic forms cause tumors in the upper digestive tract and bladder and an acute form causes hemorrhagic diathesis [1-5]. The entire plant is toxic; however, during budding, the concentration of toxic principles increases and the toxicity is maintained even when the plant is desiccated [1]. The toxic components are cyanogen glycosides (the main compound is prunasin), thiaminase (an enzyme that can cleave and inactivate thiamine), and the major carcinogenic compound ptaquiloside [6,7].

According to Anjos, et al. [3], ingestion of doses greater than 8 g/kg of the plant for more than 53 days can induce acute intoxication in cattle. The authors observed that 47 to 52 days elapsed from the time of ingestion to the appearance of clinical signs. Bovines exhibit signs of high fever, hemorrhagic lesions in different organs, bloody feces, and alterations in the heart and respiratory rates [2-4].

When ingested in a sufficient quantity to induce the acute condition, the effect of the plant is directly on the bone marrow, causing depression of hematopoietic activity and aplasia [8]. Anjos, et al. [2] studied bovines with acute

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clinical conditions, and their hematological evaluation showed leukopenia with neutropenia and thrombocytopenia and morphological evaluation showed platelets of normal size and structure.

In ovine species, Evans, et al. [9] reported the occurrence of a neurological condition, caused by thiaminase, after the inclusion of fern rhizomes in the diet. In addition, Barnett, et al. [10] described the appearance of a degenerative retinal lesion in ewes that had consumed fern for more than 150 days. However, existing literature does not present information on the acute toxic effect of this plant in sheep. This study aims to report the hematological profile and clinical information of sheep that received *P. aquilinum* buds experimentally.

This study was approved by the Unoesc Animal Experimentation and Ethics Committee - CEUA/UNOESC, protocol number 24/2013. For the experiment, healthy, mature, and mixed breed wethers were tested with green fern buds. The animals were identified as "Ovine 1" (76 kg) and "Ovine 2" (57 kg) and were kept in  $3 \times 3$  m<sup>2</sup> stalls in the Veterinary Hospital of Unoesc (West University of Santa Catarina), Xanxerê. Ten days before the experiment, the animals were placed in the stalls to adapt to the environment. Furthermore, the presence of parasites (egg count per gram of feces) and physiological clinical conditions of the animals were monitored, minimizing any interference with the results.

The diet provided to the animals consisted of corn silage with *P. aquilinum* sprouts added in daily doses of 8.5 g/kg for Ovine 1 and 11.8 g/kg for Ovine 2, representing 1.5% of the live weight of the animals. Diets were provided at three different periods during the day: morning, early, and late afternoon. The fern buds used in the experiment were harvested, crushed, and supplied fresh along with the silage. The diet was provided to the animals for 16 days, totaling 10,310 g for Ovine 1 and 10,760 g for Ovine 2.

The animals were monitored and submitted for a clinical examination twice a day. The parameters evaluated included ruminal motility, heart and respiratory rates, rectal temperature, oral and ocular mucous membrane color, and capillary refill time. To evaluate the hematological profile, eleven blood samples were taken before the investigation began, and throughout the experiment; blood samples were collected from the animals by jugular puncture using vacuum tubes with an anticoagulant (ethylenediaminetetraacetic acid). The samples were sent to the Clinical Pathology Laboratory in the Veterinary Hospital of Unoesc to perform a complete blood count. Total erythrocytes, leukocytes, platelets, and hemoglobin measurements were performed using an automated hematology counter (ABX Micros ESV60-Horiba).

No changes in clinical or hematological parameters were observed during the experiment. After the ingestion of fern, the animals were monitored for 4 weeks through a complete blood test and clinical evaluation and no clinical-pathological changes were observed. The leukogram and platelet values (Tab. 1) were consistent with the data of Anjos, et al. [3] that correspond to the cell lineage affected in acute cases of fern intoxication.

In an experiment with five cattle, Naftalin, et al. [8] observed a significantly reduced platelet count, while the leukocyte count was slightly low. In the bone marrow, there was a decrease in cellularity, presumably owing to inhibition of the formation of blasts. Furthermore, medullary aplasia was observed, and this change was observed between the 19th and 27th days after the ingestion of fern, although there was no clinical signs of intoxication [8]. Anjos, et al. [3] reported that doses above 8 g/kg are toxic if ingested for a minimum period of 53 days. Therefore, the time of ingestion is a possible explanation for the fern not having a toxic effect.

Tokarnia, et al. [4] reported that daily doses greater than 10 g/kg in cattle, when ingested for a few weeks, can cause the acute form of the disease. However, hemorrhagic diathesis begins 3 weeks after ingestion, but the animals in this experiment were monitored for longer than 3 weeks and showed no hematological changes, which explains the absence of clinical signs.

Most field cases in the bovine species are reported between September and December [3]. Marçal, et al. [1] reported cases of intoxication in July after the plant sprouted under intense frost conditions. In this study, the fern was harvested and supplied in August, and this may have influenced the lack of clinical signs, suggesting that during this period, the sprouts could be less toxic.

The absence of changes in the leukocyte and platelet counts in the tested animals suggests that the fern is not toxic to ovines at the doses provided (8.5 and 11.8 g/kg), during the period of ingestion (16 days) evaluated and at the time of plant development (August). There is a need to evaluate the effect of the fern at higher doses and for longer periods of ingestion, alongside harvesting the plant in the budding phase when the effect of its toxic principles is more intense. Finally, there is a need to test sprouting at the other times of the year as well.

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