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Genotoxity of nitroxynil and moxydectin in sheep

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ABSTRACT

Adam ML, Torres RA, Sponchiado G, Requião L, Oms PA Genotoxity of nitroxynil and moxydectin in sheep, Online Journal of Veterinary Research 9 (2) :84-87, 2005. Micronuclei assays were used to determine the genotoxicity of nitroxynil (Devoxin®) and moxidectin (Cydectin®) given 1ml/50kg and 1ml/25kg at 45 and 105 days of age, respectively, in 2 groups of 5 Suffolk sheep. There was no significant difference between treated and control (untreated newborn) animals in the frequency of micronucleated cells.

KEY WORDS: micronucleus assays, genoxicity, anthelmintics, sheep

INTRODUCTION

Peripheral blood micronucleus (MN) frequency analysis is an indication of structural and numerical abnormalities in DNA induced by chemical agents (Mavounin et al 1990; Shelby 1993), mutagens (Tucker and Preston 1996) and also by veterinary medical products (Woodward 2005). The potential genotoxicity of the antiparasite compounds nitroxynil (3-iodine, 4 hydroxide, 5 nitrobenzotrinil; (Dovenix) and moxidectin (Cydectin) was evaluated in sheep by micronucleus (MN) frequency analyses.

MATERIAL AND METHODS

Female Suffolk sheep from The Canguiri Experimental Station, Ovinoculture Section, Federal University of Paraná, in Curitiba, in Paraná, Brazil were used. Two groups of 5 sheep each were given 2% Cydectin (nitroxynil) and 4% Dovenix (moxidectin) at the rate of 1ml/50kg and 1ml/25kg at 45 (1st dose – sample I) and 105 (2nd dose – sample II) days of age. Venous blood (5ml) was collected into heparinized syringes one week after treatment. Blood smears were made on sterile, dry slides, fixed with absolute methanol and maintained at room temperature for 24 hours. Slides were then immersed in giemsa solution (1ml of giemsa in 30ml phosphate buffer with pH = 6.8) for 10 minutes.

One thousand cells were analyzed per sheep. The number of micronucleated cells was counted using an optical microscope with oil-immersion lens. Analysis of variance (ANOVA) was used to compare the rate of micronucleated cells among treatments, using Statistica v.6.0 (Statsoft Inc.).

RESULTS

While the frequency of micronucleation varied among treatment groups (Control = 2.12 x 10^{-3}, 45 days = 3.96 x 10^{-4} and 105 days =3.33 x 10^{-3}; Tables 1, 2 and 3), the differences were not significant (ANOVA, p 0.32; Figure 1).

Table 1. Summary of the cells sampled in the Control group. AR= animal registry; NC= normal cells; MN= micronuclei; TC= total of cells.

AR	NC	MN	TC
341	1027	09	1036
331	1000	00	1000
938	1000	01	1001
313	1000	01	1001
337	1137	00	1137
Total	5164	11	5175
Frequency	2.12 x 10^{-3}		

Table 2. Summary of the cells sampled in the 45 days group. AR= animal registry; NC= normal cells; MN= micronuclei; TC= total of cells.

AR	NC	MN	TC
897	1020	00	1020
907	1024	00	1024
930	1000	01	1001
903	1006	01	1007
940	1003	00	1003
Total	5053	02	5055
Frequency	3.96×10^{-4}		

Table 3. Summary of the cells sampled in the 105 days group. AR= animal registry; NC= normal cells; MN= micronuclei; TC= total of cells.

AR	NC	MN	TC
331	1013	07	1020
306	1023	07	1030
350	1020	00	1020
341	1030	00	1030
307	997	03	1000
Total	5083	17	5100
Frequency	3.33×10^{-3}		

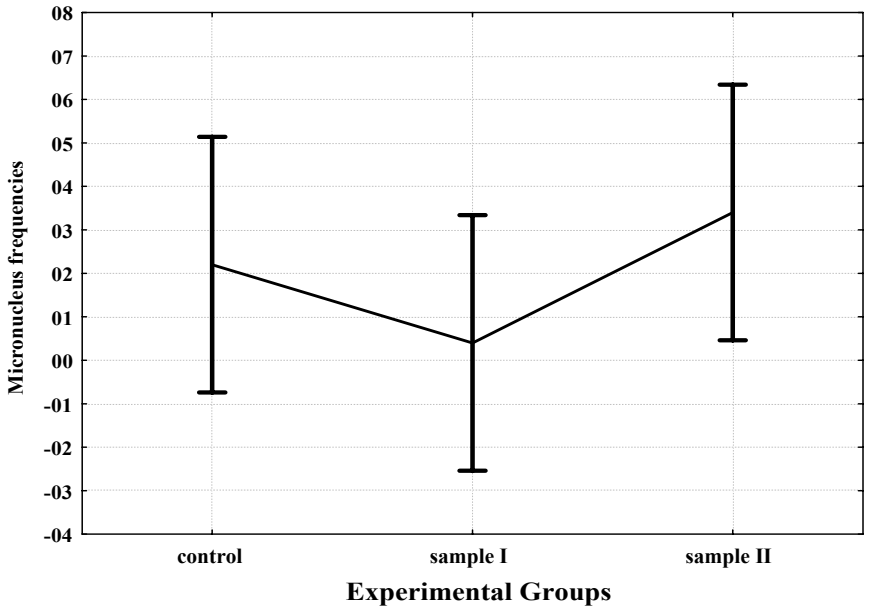

Figure 1. *One-way* ANOVA results (p=0.32) for micronucleic frequencies. Vertical bars denote 0.95 confidence intervals.

DISCUSSION

Controls had a spontaneous basal rate of micronucleation (2.12 x 10^{-3}), comparable with background micronuclei frequencies shown to be around 2 x 10^{-3} in other species and 0.69 x 10^{-3} in *Ovis aires* (Mavournin et al 1990; Cristaldi et al 2004). The findings show no relationship between the use of Dovenix and Cyndectin and appearance of micronuclei.

The detection of genotoxicity due to chemical agents is related with the number of cell cycles exposed to those agents (Miller et al 1993, Lohmann 1995). In our experiments the blood samples were taken 1 week after treatment, suggesting that the cells may not have micro-nucleated. On the other hand, the time between treatments may also have caused DNA repair to mask effects of treatment (Alberts et al 1997).

The 2nd dose (Table 2; Figure 1) was expected to increase micronucleation. However, the results obtained herein suggest that there is no putative chronic genotoxicity of the chemical components (3-iodine, 4 hydroxide, 5 nitrobenzotrinil and moxidectin). A possible explanation would be that repeated treatments result in steady-state levels of micronucleated cells. The low frequencies of micronuclei suggested that extended exposures reduced micronucleus response compared with acute exposure (MacGregor et al 1990).

In most species, micronucleated blood cells are removed from the peripheral blood stream by the spleen (MacGregor et al 1990), and the spleen may remove micronucleated cells very efficiently, resulting in the low observed frequency (Table 1 and 2).

A comparison of the Control group and the 45 day group (sample 1), showing an order of magnitude decrease in micronucleated cells at 45 days (Fig. 1, Tables 1, 2), could suggest high toxicity occurring in the bone marrow, causing a suppression of the blood cell development. Similar patterns of decreasing of micronucleated cells as well mitotic index were observed by Montero and Ostrosky (1997) studying the genotoxicity of Praquizantel.

While this last possibility could imply leucopoenia, our results suggest that no genotoxic effects occur with the recommended dosage of moxidection and nitroxynil in sheep

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