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# Transfer of Ochratoxin A into ewe's milk following a single or chronic ingestion of contaminated feed

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## INTRODUCTION

Ochratoxins, a group of highly toxic metabolites produced by some species of *Aspergillus* and *Penicillium*, are commonly found in foods and animal feeds. Ochratoxin A (OTA), the most important toxin of this family, is nephrotoxic, hepatotoxic, teratogenic and carcinogenic in animals and was recently classified by the International Agency of Research on Cancer (IARC) as a class 2B, possible human carcinogen. Ruminants, thanks to the action of rumen microorganisms, have the capacity to hydrolyze OTA into the less toxic compound ochratoxin  $\alpha$  (OT $\alpha$ ). However, not all the ingested toxin is degraded in the rumen. The undegraded OTA is absorbed in the gastrointestinal tract and can contaminate animal products posing a potential risk for consumers. Although the presence of OTA has been reported in bovine milk, the transmission from contaminated feed into ruminants' milk is not well known. In this study the effect of a single or chronic OTA ingestion on the transmission of the toxin into milk was examined in dairy ewes.

## MATERIALS AND METHODS

- Six dairy ewes (71.3  $\pm$  7.9 Kg) in late lactation were randomly divided in two lots that received 5 (Dose 1) or 30  $\mu$ g (Dose 2) OTA/kg body weight/day. Animals were fed at libitum a diet consisting of 28 % wheat-based concentrate and 72% hay twice a day. Contaminated wheat that was experimentally inoculated with a toxigenic *Aspergillus ochraceus* strain was the toxin source. OTA-contaminated ground wheat was orally administered as a bolus once a day before the morning feeding to ensure that animals consistently received the desired quantity.

- The single doses were administered on day 1. Plasma and milk samples were collected on day 2. Then, chronic administration started on day 5 and lasted for 28 days. Samples were collected during the steady state period at days 6, 9, 15, 22 and 28.

- Plasma and milk concentrations of OTA were determined by high-performance liquid chromatography (HPLC) with fluorescence detection. Prior to HPLC analysis, samples were extracted with chloroform and purified using immunoaffinity columns.

## RESULTS

- For both doses, OTA was detected in plasma and milk, indicating that part of OTA was not degraded by the ruminal microbiota.

- The transfer of OTA into milk was dose dependent. Chronic administration did not increase the concentration of OTA in milk (Table 1 and 2).

- OTA concentration in milk following daily administration remained relatively stable from 1 week up to the end of the 4-week experimental period.

- OTA carry-over rate was low and did not vary between single or chronic administration (Table 1).

- Concentration of OTA in plasma and milk varied widely among animals (Table 2).

Table 1: OTA carry-over into ewe's milk in relation to OTA intake.

	OTA concentration in milk (ng/L) (a)	Carry over rate (%) (a)
<b>Single administration</b>		
5 $\mu$ g.Kg <sup>-1</sup> .day <sup>-1</sup>	87.5 $\pm$ 84.2	0.037 $\pm$ 0.032
30 $\mu$ g.Kg <sup>-1</sup> .day <sup>-1</sup>	315.1 $\pm$ 292.1	0.032 $\pm$ 0.046
<b>Chronic administration (28 days)</b>		
5 $\mu$ g.Kg <sup>-1</sup> .day <sup>-1</sup>	43.4 $\pm$ 23.1	0.013 $\pm$ 0.008
30 $\mu$ g.Kg <sup>-1</sup> .day <sup>-1</sup>	323.3 $\pm$ 200.7	0.019 $\pm$ 0.007

(a): (Mean  $\pm$  SD, n=3)

Table 2: milk:plasma ratio of OTA during the steady state

Ewe	Day	Plasma concentration ( $\mu$ g/L)	Milk concentration ( $\mu$ g/L)	Ratio (milk/plasma)
<b>5 <math>\mu</math>g.Kg<sup>-1</sup>.day<sup>-1</sup></b>				
1023	6	0.760	0.033	0.04
	15	0.565	0.026	0.05
	22	0.554	0.029	0.05
	28	0.450	0.047	0.11
2051	6	0.725	0.033	0.05
	15	0.606	0.028	0.05
	22	0.560	0.043	0.08
	28	0.210	0.027	0.13
<b>30 <math>\mu</math>g.Kg<sup>-1</sup>.day<sup>-1</sup></b>				
377	6	4.253	0.276	0.06
	15	3.992	0.222	0.06
	22	7.356	0.182	0.02
	28	1.411	0.112	0.08
2065	6	5.613	0.750	0.13
	15	10.848	0.626	0.06
	22	2.984	0.618	0.21
	28	3.000	0.621	0.21

## CONCLUSIONS

- OTA at concentrations that can be present in naturally contaminated feeds, escape ruminal degradation and can be found, although in low quantities, in ewe's milk.

- The conversion of OTA to OT $\alpha$  is incomplete. Additional investigations are necessary to determine the maximum level of OTA that the ruminant can detoxify.

