A National Survey of Mycotoxins in Canada

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ABSTRACT

Mycotoxin contamination of animal feedstuffs continues to be an area of concern in various regions of Canada. A survey of research institutions across Canada revealed that the mycotoxins, zearalenone, ochratoxin A, tricothecenes and aflatoxin, have been detected in suspected feed samples. These mycotoxins have produced mycotoxicoses in poultry, hogs and cattle. Estrogenic syndromes in hogs may be caused by the ingestion of zearalenone produced by *Fusarium* molds on grain corn grown in southern Ontario. Overwintered barley contaminated with T-2 toxin was suspected in an outbreak of fusariotoxicoses in swine in northern Alberta. Central and eastern Canada, which seem to be the major focus of mycotoxin occurrence, will be the object of further field level examination to discover patterns of occurrence and economic impact.

INTRODUCTION

The most frequent types of mycotoxins occurring in Canada appear to be produced primarily by *Fusarium* species. Zearalenone or F-2 toxin is produced commonly on corn and is attributed mainly to *Fusarium roseum* and *Fusarium* graminearum (1-4). *Fusarium* species produce other important toxins, including T-2 toxin as well as other trichothecenes (5). Fungi are often associated with corn which is immature or high in moisture at time of harvest. The molds develop under cool, wet weather conditions, especially during the autumn and winter in the field or in storage.

It has been known for some time that the cool climate existing in Canada provides toxin-producing Penicillia (P. viridicatum and P. palitans) with a competitive advantage (6). This is illustrated by the presence of ochratoxin A and citrinin in cereal grains (wheat, rye, barley and oats) which were overwintered in the field due to wet weather at harvest time in the autumn.

Aspergillus species are less common in Canada, although they do occur. A. ochraceus produces ochratoxin A (7,8). A. flavus and A. parasiticus produce various aflatoxins. These fungi occur predominantly in areas of high temperature (+20 C) and high humidity. Aflatoxins do not often occur naturally in Canada. Aflatoxins could possibly be a problem for livestock and humans who consume products such as peanuts and cottonseed meal imported from warm or humid countries.

The prime mycotoxicosis associated with mycotoxin occurrence in Canada appears to be the estrogenic syndrome in hogs associated with the ingestion of zearalenonecontaminated corn. Another important aspect in hogs and cattle is feed refusal, vomiting, loss of weight and internal hemorrhaging, which is probably linked to the trichothecene vomitoxin. In addition, T-2 toxin and, occasionally, ochratoxin may produce mycotoxicosis in poultry and cattle.

There was a need to establish what kinds of problems were experienced by livestock producers across Canada. In this paper, the authors present data collected as part of a national survey of mycotoxins and mycotoxicoses.

METHODS

In 1978, the Animal Research Centre of Agriculture Canada began a study to determine the extent and nature of mycotoxin occurrence in Canada. The consulting firm of Deloitte Haskins & Sells Associates was contracted to complete a national survey. A survey of government agencies at the federal and provincial levels, as well as universities and private companies, began during spring 1979.

The country was divided into 3 regions: British Columbia and the prairies in the west, Ontario in the center, and Quebec and the Atlantic provinces on the eastern coast. Interviews with government agencies were designed to produce the following information: (a) types of research being conducted on analytical methods or animal toxicology; (b) methods of analysis currently being used for mycotoxin testing; (c) types of mycotoxins present in various regions of Canada; and (d) types of mycotoxicoses resulting from mycotoxin contamination.

RESULTS

British Columbia and the Prairies

A provincial government laboratory in British Columbia has been testing for mycotoxins since 1970 on a demand basis. About 200 samples were analyzed with 21 positives for T-2 toxin and 1 positive for ochratoxin A.

In 1976, Puls and Greenway (9) documented a problem with T-2 mycotoxicoses. The outbreak caused mortality in geese related to upper alimentary and intestinal necrosis, and upper alimentary distress occurred in horses and swine. The suspected grain was barley; thin layer chromatography (TLC) indicated a T-2 level of 25 ppm. There have been no additional positive mycotoxin tests from this provincial laboratory during the past 5-6 years.

In Alberta, mycotoxin analyses have been done by a provincial government laboratory. Of 100 samples which have been analyzed since 1973 (A. Strausz, unpublished data), only a few of the samples were positive at trace levels: 2 aflatoxin, 3 sterigmatocystin, 3 ochratoxins and 3 F-2 toxin (zearalenone). The laboratory used TLC and a mass spectrometer in their early analysis. At present, a gas chromatograph-mass spectrometer is used.

An outbreak of mycotoxicosis in swine occurred in 1975 in the Peace River region of Alberta. Analysis of the barley involved, which had been overwintered or harvested at high moisture levels, revealed a positive bioassay for T-2 toxin. Chemical analyses for T-2 were negative; however, feeding trials with hogs, chickens and geese produced symptoms consistent with T-2 toxin ingestion (10-12). The hog trials showed a large refusal factor, necrosis in the mouth and poor growth. The chickens and geese exhibited severe oral lesions and when force-fed, developed severe lesions of the esophagus.

A second Alberta testing laboratory located in Calgary has been using TLC for mycotoxin testing. The laboratory received samples from across western Canada. In 1978 and 1979, they tested 271 samples for aflatoxins, ochratoxins, T-2 toxin and zearalenone. The only positive mycotoxin test was in a corn sample from Manitoba in which a 1-ppm level of zearalenone was detected.

In Saskatchewan, mycotoxin analyses have been conducted regularly by the Agriculture Canada laboratory in Saskatoon (13). The laboratory used TLC to characterize toxins (14-18), except the trichothecenes, for which a bioassay (guinea pig) was used (19). During the 5-year period 1971-75, 213 samples were analyzed for myco-toxins. The results were: (a) 1 positive for aflatoxin B, alfalfa hay (50 ppb) (b) 7 positive for ochratoxin A, 6 wheat (30-6,000 ppb) and 1 hay (30 ppb).

In Manitoba, the Provincial Diagnostic laboratory in Winnipeg has detected 3 positive for T-2 toxin using a bioassay method during the past 5 years. The Agriculture Canada Research Centre at the University of Manitoba in Winnipeg is conducting mycotoxin research including improvement of analytical methods, analysis of suspected feedstuffs and monitoring of mycotoxin occurrence at the field level (J.T. Mills and D. Abramson, unpublished data). Preliminary work indicates the presence of ochratoxins and T-2 toxin in a small number of samples in southern Manitoba and the prairies. This laboratory uses TLC (20) for mycotoxin screening and high pressure liquid chromatography (HPLC) for aflatoxin and ochratoxin testing.

Ontario

At present, data suggest that the occurrence of mycotoxins may be most prevalent in Ontario, particularly in the more humid southern areas such as the Niagara peninsula. Testing has been done by the Veterinary Services Branch of the Ontario Ministry of Agriculture and Food in Guelph, Ontario. A summary of analytical results on suspected samples for 1972-77 is presented in Tables I and II (21). The primary mycotoxin detected was zearalenone at an average level of 3.85 ppm with a range of 0.01-141 ppm. Twenty percent of samples was between 1 and 10 ppm; 3% was between 10 and 100 ppm. The highest aflatoxin level was 0.8 ppm. The analytical method for screening mycotoxins was TLC (18). The predominant mycotoxin detected was zearalenone, which occurred in corn infected with Fusarium. The primary mycotoxicosis related to Ontario mycotoxins was vulvovaginitis or the estrogenic

TABLE I

Number	of Specim	ens Positive	for Individual	Mycotoxins in
Various	Suspected	Fcedstuffs :	for 1972-77	•

syndrome in hogs. This disease is characterized in young gilts by swelling and edema of the vulva, enlargement of the uterus and general reproductive disorders (23,24).

Quebec and the Atlantic Provinces

The Quebec Provincial Government laboratory at St. Foy is equipped to test for mycotoxins. At the time of the survey, there had been no positive mycotoxin analyses at the laboratory for the previous 2 years.

Studies at the Nappan, Nova Scotia, Research Station of Agriculture Canada have identified a fungal metabolite of soil-borne fungi which causes "ill-thrift" in livestock. The research suggested that an antibiotic produced from the fungi reduced rumen flora in sheep (25). The fungi have been identified as *Chaetomium cochliodes* and *C. globosum* and the toxic metabolites characterized as chetomin (26). The toxicity of chetomin (27) was examined with bacteria and animals. The toxin was found to be bacteriostatic against several gram-positive bacteria with little activity against gram-negative organisms.

In rats, a dose of 50 ppm caused weight reductions of 50% and a 100-ppm dose resulted in death after 4-6 days. Hematuria and hemorrhage in the lumen of the gastrointestinal tract were common in the lethal-dose group. Sheep (33-kg), when dosed with chetomin at 1 ppm for 5 days, lost 0.5 kg in 10 days while the control gained 0.5 kg. Ewe lambs receiving 5 ppm or more showed immediate symptoms and died within 24 hr; they were observed to have acute peritonitis and subindocardial hemorrhages (28).

There are no other laboratories testing for mycotoxins in the Atlantic provinces.

DISCUSSION

The survey illustrated that the awareness and incidences of mycotoxin occurrence varies widely across Canada. In general, the cool climate restricts the range of fungi and

Mycotoxin	1972	1973	1974	1975	1976	1977	
	ΑΕΟΤΖ	ΑΕΟΤΖ	ΑΕΟΤΖ	ΑΕΟΤΖ	AEOTZ	ΑΕΟΤΖ	
Corn	30	4	1 11	1 1 17	1 - 1 - 18	2 4	
Mixed feed				37			
Grain	-			4			
Other Total sampled	209	219	256	1 500	1 347		

Source: Funnell 1979 (21).

A = Aflatoxins, E = ergot, O = ochratoxins, T = T-2 toxin, Z = zearalenone,

TABLE II

Comparison between Specimens Containing One or More of the Mycotoxins and Those Containing Zearalenone

	1972	1973	1974	1975	1976	1977
Total specimens Specimens with one	209	219	256	500	347	491
or more mycotoxin(s)	35	9	17	113	38	65
Positive (%)	16.7	4.1	6.6	22.6	11.0	13.2
Specimens with zearalenone	34	9	14	112	33	64
Positive with zearalenone (%)	16.3	4.1	5.5	4	9.5	13.0

Source: Funnell 1979 (21).

Positive zearalenone samples-levels from 0.01 ppm-141 ppm; 55 samples-1 ppm-10 ppm; 8 samples-10 ppm; 3 samples-over 100 ppm.

toxins present in Canadian feeds.

The majority mycotoxin occurrence in western Canada, British Columbia and the prairies has been in cereal grains. Contamination occurs when these crops receive excess moisture during harvest, or are overwintered and harvested in the spring. T-2 toxin, primarily from Fusarium tricinctum, and ochratoxin A from Penicillium viridicatum and P. palitans are most prevalent under these conditions and have caused the only documented cases of mycotoxicoses in western Canada,

The Peace River area is predisposed to mycotoxin development. The short growing season in certain areas, 90-110 frost-free days, and frequent rains during September and October create ideal conditions for mold growth and mycotoxin development. Since home-grown cereals are frequently fed to farm livestock, contaminated grain would not be diluted before consumption. Dilution is more likely to occur when grain is transported to feed mills and mixed with other feeds. The most severe documented outbreak of mycotoxicoses in western Canada occurred in the Peace River area.

Southern Manitoba, particularly in the Red River Valley, is a moist, humid region which supports significant corn production. A small level of occurrence of most mycotoxins has been detected; ochratoxin A is the most common. As the corn industry develops, it is anticipated that the fusariotoxins such as zearalenone will become more common.

Ontario is the region of Canada that probably has the highest level of mycotoxin occurrence. Research and testing are also more advanced there than in other regions of Canada. The presence of zearalenone in corn has created new management practices in the hog industry in southern Ontario. Producers are reluctant to feed home-grown corn to their breeding sows and gilts because of the estrogenic effects of zearalenone. This disrupts the total integration of these farms and forces the purchase of grain from unaffected areas such as western Canada, which has no history of zearalenone contamination.

Quebec and the Atlantic provinces which have many areas with moist, humid conditions have not reported a significant occurrence of mycotoxins. In fact, the occurrence of vomitoxin in Quebec in 1980 was the first significant feed grain toxin for many years. The Atlantic provinces have also had few positive mycotoxin results although field practitioners, particularly in poultry, are aware of mycotoxicoses in their diagnoses.

The majority of persons interviewed, whether or not they were in an area where mycotoxin occurrence was common, felt that the lack of systematic methods for reporting of mycotoxin occurrence severely restricts accurate analysis of the extent of the problem: Another major factor which prevents accurate analysis of mycotoxins and mycotoxicoses is the lack of standardized analytical procedures. It was stated that the poor sensitivity of existing tests limits the accurate diagnoses of mycotoxicoses. The small number of labs at which samples can be analyzed affects the turn-around time of test results and thus precludes mycotoxicoses consideration in many diagnoses.

Another major concern was the lack of research information available on toxicity levels of various mycotoxins for various ages and types of commercial livestock. Mycotoxicosis is not well understood in many areas and, although many researchers feel there is some type of problem related to fungal metabolites, they do not have the resources to adequately define the extent of the problem in their area.

There is concern among researchers that mycotoxins may enter the food chain, either directly, in plant products, or indirectly, as residues in meat products, such as meat, milk or eggs. More research is necessary to establish the potential health hazards associated with mycotoxin contamination.

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