

Reprinted from

Journal of Food Protection Vol. 41, No. 5, Pages 399-403 (May, 1978)

Copyright © 1978, International Association of Milk, Food, and Environmental Sanitarians

Trichothecenes: Occurrence and Toxicoses

A. CIEGLER

Northern Regional Research Center, Agricultural
Research Service, U. S. Department of Agriculture,
Peoria, Illinois 61604

(Received for publication August 10, 1977)

ABSTRACT

The Δ^9 -12,13-epoxy-trichothecenes are a group of 37 naturally occurring sesquiterpenoids produced by species of *Fusarium*, *Stachybotrys*, *Myrothecium*, *Trichoderma*, and *Cephalosporium*. They are responsible for a variety of mycotoxicoses in man and animals that may be manifested by severe dermatitis, vomiting, bloody diarrhea, decreased weight gains, extensive hemorrhaging, and death. Outbreaks, which have occurred in Russia, Europe, the United States, Japan, and Korea, usually result after cereal grains in the field have been exposed to prolonged cool and wet weather. Hence, these mycotoxicoses do not appear to be a problem of faulty grain storage, since they involve field fungi primarily rather than storage molds.

The trichothecenes are a family of about 37 naturally occurring sesquiterpenoids produced by various species of fungi. They are derivatives of the trichothecane ring system (Fig. 1a) named after trichothecin, the first member of the group to be isolated. All of the naturally occurring toxins contain an olefinic bond at 9, 10 and an epoxy group at 12, 13 and, therefore, may be characterized as 12, 13-epoxy-trichothecenes (Fig. 1b). In addition, most of these toxins have at least one $-\text{OH}$ or ester group at position 4 as in Fig. 1b. The compounds can be subdivided into groupings based on substitution at position 8 and the presence (or absence) of a macrocyclic ring (Fig. 2-4).

Additional subgroupings are possible, e.g., the presence or absence of an epoxide between positions 7 and 8 as in crotochin and crotochol.

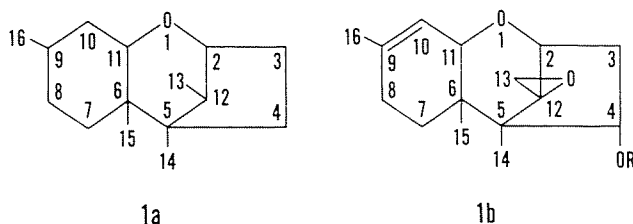


Figure 1. Basic ring structures of trichothecane and 12,13-epoxy-trichothecenes.

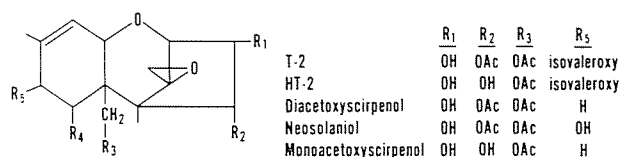


Figure 2. Structures of 8-keto trichothecenes.

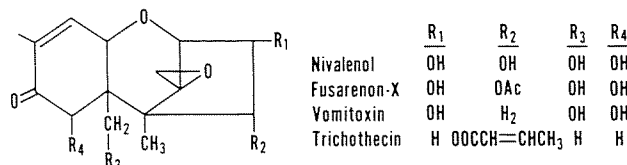


Figure 3. Structures of trichothecenes substituted at position 8.

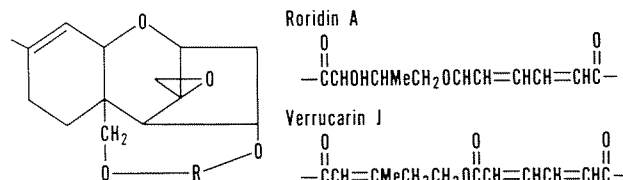


Figure 4. Structures of macrocyclic trichothecenes.

FUNGAL SOURCES

The 12,13-epoxytrichothecenes are produced by members of a number of fungal genera including *Fusarium*, *Stachybotrys*, *Myrothecium*, *Trichoderma*, and *Cephalosporium*; but pragmatically, with respect to serious contamination of food or feedstuffs, we will be concerned primarily with members of the genera *Fusarium* and *Stachybotrys*. Considerable confusion exists concerning the taxonomy of the fusaria, and I am not qualified, nor is this the time and place, to discuss this aspect; I will use, in general, the simplified nomenclature of Snyder and Hansen who recognized only nine species of *Fusarium* (26).

TOXICITY

The 12, 13-epoxytrichothecenes are toxic to animals, plants, insects, fungi, protozoa, tumor cells, and cultured cells. However, as a class they are rather poor antibiotics. In humans and animals, many of these compounds can cause a severe dermatitis, vomiting, bloody diarrhea, decreased weight, extensive hemorrhaging, and death. Pathological lesions in animals include cellular damage and karyorrhexis to the proliferating tissues of intestinal mucosa, bone marrow, spleen, testis, and ovary. On a molecular level, they may cause inhibition of protein and DNA synthesis. The LD₅₀ of some select trichothecenes is shown in Table 1.

TABLE 1. LD₅₀ of select trichothecenes

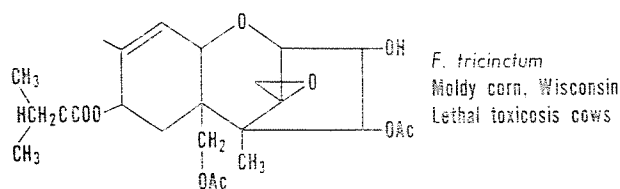
Toxin	Mice (mg/kg)
Fusarenon-X	3.3
Nivalenol	4.1
Vomitoxin	50.0
T-2	5.2
HT-2	9.0
Diacetoxyscirpenol	23.0
Trichothecin	< 250
Verrucaric A	0.5-0.75

INCIDENCE

What is the incidence of trichothecenes in agricultural commodities, and what role do these compounds play in mycotoxicoses? These questions should be separated from the incidence of fungi capable of producing trichothecenes that can be isolated from various commodities. Put differently, it is a question of direct vs. circumstantial evidence and the two are often confused in the literature. Direct evidence, particularly that also implicating a mycotoxicosis, is often extremely difficult to obtain.

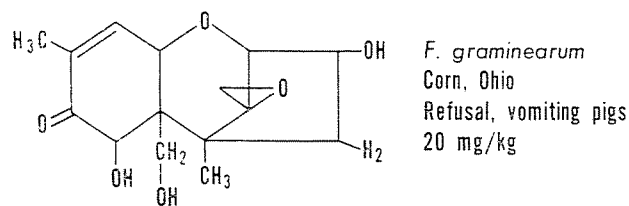
To the best of my knowledge, there are only a few documented reports of trichothecenes actually being detected in contaminated commodities. These are summarized in the following figures (Fig. 5-8).

Although only a few reports have appeared in the literature, considerable circumstantial evidence exists to link *Fusarium*-contaminated food and feeds to a variety of mycotoxicoses. *Fusarium* contamination of commodities is common throughout the world, particularly in geographical regions subject to cool wet periods. Intoxications of man and/or animals ascribed to consumption of *Fusarium*-contaminated commodities have been reported extensively over a long time span,



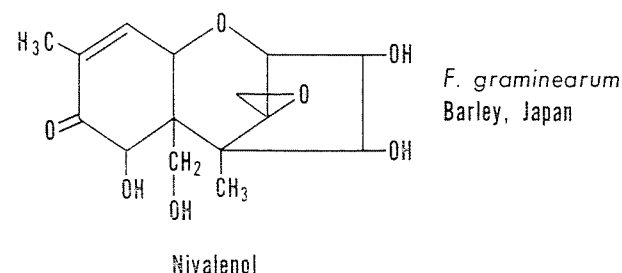
Hsu et al [9]

Figure 5. Natural occurrence of T-2 toxin.



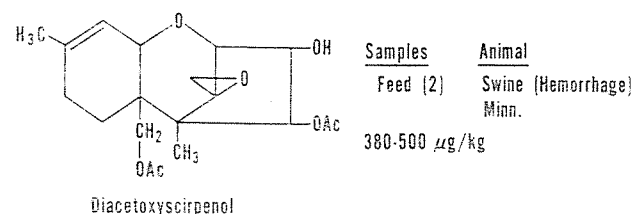
Vesonder et al [32, 33]

Figure 6. Natural occurrence of vomitoxin (deoxynivalenol).



Morooka et al [19]

Figure 7. Natural occurrence of vomitoxin and nivalenol.



Vomitoxin
Corn, mixed feed (6) Midwest
Refusal, vomiting, pigs, dogs
Conc. 50-1800 µg/kg

T-2
Mixed feed (1)
Cows, bloody stools
Nebraska
76 µg/kg

Miracha et al [17]

Figure 8. Natural occurrence of trichothecenes in feedstuffs.

particularly in Japan, Korea, Russia, and the United States. The best known of these intoxications was the outbreak of alimentary toxic aleukia (ATA) in Russia during WWII involving thousands of people; however, cases had been recorded in Eastern Siberia as early as 1913. The disease was ascribed to toxic steroids in *Fusarium*-contaminated grains, but it was later shown that one of these toxic steroid preparations actually contained 12, 13-epoxytrichothecenes. Yagen (38) offered compelling evidence that T-2 toxin produced by *Fusarium poae* and *Fusarium sporotrichoides* (*Fusarium tricinctum*) was the causative compound.

Another aspect of fusariotoxicoes prevalent in the United States involves the refusal-vomiting phenomena affecting pigs fed corn infected with *Fusarium graminearum*. Vomiting and nausea have also been reported among humans in Japan from consuming *Fusarium*-

contaminated rice or wheat products (23) and in Russia from eating bread baked with scabbed wheat (4,20). The literature is extremely confusing on the causative toxin; various investigators have shown that a number of trichothecenes can elicit vomiting in laboratory animals. I suspect that most trichothecenes can cause vomiting on ingestion. However, this is not the point; the question is rather which toxins actually occurring in the field are causing toxicoses; which are major, which are minor; do they interact? To date, definitive experiments to clarify all these questions have not been carried out.

The refusal-vomition phenomenon has been described as caused by *F. poae* which produced T-2 toxin (5,15) or by presence of fusaranone-x or nivalenol (29,30). Vesonder and his colleagues (32,33) at our laboratory and later Ishii et al. (10) have definitively shown that *F. graminearum* which produced vomitoxin (deoxynivalenon) in corn was the cause of both refusal and vomiting. Morooka et al. (19) earlier had isolated vomitoxin from contaminated barley but had not characterized it. The most recent outbreak of fusariotoxicosis on large scale was in 1972 and involved corn in a region stretching from Pennsylvania to Nebraska. FDA personnel analyzed 223 samples of this corn (7), and 93 of 173 samples contained a skin irritant by the rabbit skin test. We have started an analysis of 10 of these samples and have found vomitoxin and T-2 in some; these assays are continuing.

Previous outbreaks of fusariotoxicoses of cereal grains in the United States which caused intoxication in swine occurred in 1928, 1957, 1958, 1964, 1965; an outbreak in 1963 caused nausea, vomiting, abdominal pain, and diarrhea in humans (3).

T-2 toxin produced by *F. tricinctum* has received most attention among the trichothecenes for a variety of reasons; mycotoxicoses believed caused by a trichothecene are often ascribed to this compound, even without evidence. However, in the United States *F. graminearum*, a non T-2 producer, occurs more often than *F. tricinctum*. Nevertheless, a well-documented case where T-2 toxin caused deaths among dairy cows in Wisconsin was reported by Hsu et al. (9).

A disease syndrome in poultry and fancy pigeons (characterized by raised yellowish-white lesions in the oral cavity, on the feet and shanks, and around the eyes) observed in the Southeast could be mimicked by feeding chickens small concentrations of T-2 toxin (35-37). However, T-2 toxin was not isolated from the feed involved.

The most dramatic mycotoxicosis, probably involving T-2 toxin as well as other trichothecenes, was the previously mentioned ATA that occurred in Russia during WWII and earlier. Because of the inadequate state of the scientific art at that period, the toxic agents involved were reported to be steroids (11). More recent work has clarified the situation so that strong circumstantial evidence implicates T-2 toxin as the major toxin involved (12,16,39).

F. tricinctum capable of producing T-2 toxin and a toxic butenolide have been isolated from toxic fescue grass (8). However, the dry-type gangrene which occurred in cattle eating this contaminated forage has not been reproduced by administration of these substances.

Fusariotoxicoses have been reported throughout the world; Tables 2 and 3 partly illustrate the problem as occurring in Sweden, Korea, and Japan.

Stachybotryotoxicosis, a haemorrhagic disease of horses, cattle, and swine consuming hay or other cellulosic feeds molded with *Stachybotrys atra*, has been reported to occur extensively in Russia, in the Balkan countries, and possibly in Israel and Finland (14,18,25, 27,28,31). Experimental stachybotryotoxicosis has been developed in poultry (21,24). In addition, peasants in these areas have suffered severe vesicant effects from sleeping on contaminated hay. Eppley and Bailey (6) at FDA have found five trichothecenes produced by *S. atra* which probably account for the symptoms observed. *Stachybotrys* and stachybotryotoxicosis have recently been reviewed in extenso (22).

In addition to the preceding mycotoxicoses apparently caused by various species of *Fusarium*, *Fusarium moniliforme* on moldy corn appears to be implicated in equine leucoencephalomalacia in South Africa, Egypt, and in Louisiana, where swine as well as horses are affected (1,13). We have isolated *F. moniliforme* from toxic Louisiana corn, but we have not as yet been able to detect the presence of trichothecenes. Herein lies the crux of the overall problem—practical analytical methods for detection of trichothecenes in contaminated agricultural commodities have not been developed. It is obvious from the widespread occurrence of the fusaria and their enormous capacity for toxin synthesis that a potentially serious fusariotoxicosis problem can exist throughout the world. However, defining the magnitude of the problem awaits development of assays to permit the necessary surveys.

Those outbreaks which can attributed to trichothecenes have been dramatic. Less obvious and perhaps

TABLE 2. *Fusariotoxicoses in Sweden*

Fungus	Source	Host	Symptoms
<i>Fusarium moniliforme</i>	Oats, feed	Horse, chicken	Colic, gastric, growth loss
<i>Fusarium tricinctum</i>	Hay, feed	Sheep, cow, pig, chicken,	Gastric, respiratory, anorexia, growth loss
<i>Fusarium sporotrichiodes</i>	Hay	Cow	Respiratory
<i>Fusarium poae</i>	Hay, oats, feeds	Cow, horse, pig, chicken	Colic, gastric, anorexia, growth loss

more important are toxicoses exerting insidious effects. Boonchuvit et al. (2) showed an increased mortality in chickens dosed with T-2 toxin and later subjected to infection with *Salmonella* species; hens exposed to T-2 exhibit lowered egg production, thinner shells, and concomitant reduced feed efficiency and weight gains (34). Similar loss of feed efficiency and weight gains have been noted on farms where pigs have been fed corn contaminated with *F. graminearum*.

Another neglected aspect deserving attention is potential synergistic activity among the trichothecenes. Many fusaria and *S. atra* can produce more than one toxin, e.g., *F. tricinatum*: T-2 toxin, HT-2 toxin,

neosolaniol, diacetoxyscirpenol; *F. roseum gibbosum*: monoacetoxyscirpenol, scirpentriol; *S. atra*: satratoxin G and H, plus others; *F. nivale*: nivalenol, fusarenol, nivalenol diacetate; *F. roseum*: nivalenol, vomitoxin, monoacetoxyldeoxynivalenol. Under the circumstances, it is not surprising that the following variety of diseases described under numerous terms all turn out to be primarily trichothecene toxicoses: ATA, refusal-vomition phenomenon, haemorrhagic syndrome, moldy bean hull toxicosis, akakabibo toxicosis, cereal scab or scabby grain toxicosis, drunk bread (intoxicating bread) toxicosis, stachybotryotoxicosis, dendrochiotoxicosis (myrotheciotoxicosis), and fusariotoxicosis.

TABLE 3. *Fusariotoxicoses in Japan and Korea*

Date	Grain	Host	Symptoms
1932	Wheat	Horse, man	Vomiting, diarrhea
1946	Flour	Man	Vomiting
1950	Wheat	Man	Vomiting
1954	Oats	Horses	—
1955	Wheat	Man	Vomiting
1956	Wheat	Man	Nausea, vomiting, diarrhea, chills
1963	Wheat, oats corn, barley	Horses, sheep, man, pigs, dogs	Vomiting, diarrhea
1969	Fodder	Horses	—
1970	Wheat	Chicken	—

Fusaria isolated: *F. graminearum*, *F. nivale*, *F. poae*, *F. oxysporum*, and others.

REFERENCES

- Badiali, L., M. H. Abou-Youssef, A. I. Radwan, F. M. Hamdy, and P. K. Hildebrand. 1968. Moldy corn poisoning as the major cause of an encephalomalacia syndrome in Egyptian *Equidae*. *Am. J. Vet. Med.* 29:2029-2035.
- Boonchuvit, B., P. B. Hamilton, and H. R. Burmeister. 1975. Interaction of T-2 toxin with *Salmonella* infections of chickens. *Poult. Sci.* 54:1693-1696.
- Curtin, T. M., and J. Tuite. 1966. Emesis and refusal of feed in swine associated with *Gibberella zeae*-infected corn. *Life Sci.* 5:1937-1944.
- Dounin, M. 1926. The fusariosis of cereal crops in European Russia in 1923. *Phytopathology* 16:305-308.
- Ellison, R. A., and F. N. Kotsonis. 1973. T-2 toxin as an emetic factor in moldy corn. *Appl. Microbiol.* 26:540-543.
- Eppley, R. M., and W. J. Bailey. 1973. 12,13-Epoxy- Δ^8 -trichothecenes as the probable mycotoxins responsible for stachybotryotoxicosis. *Science* 181:758-760.
- Eppley, R. M., L. Stoloff, M. W. Trucksess, and C. W. Chung. 1974. Survey of corn for *Fusarium* toxins. *J. Assoc. Off. Anal. Chem.* 57:632-635.
- Grove, M. D., S. G. Yates, W. H. Tallent, J. J. Ellis, I. A. Wolff, N. R. Kosuri, and R. E. Nichols. 1970. Mycotoxins produced by *Fusarium tricinatum* as possible causes of cattle disease. *J. Agric. Food Chem.* 18:734-736.
- Hsu, I. C., E. B. Smalley, F. M. Strong, and W. E. Ribelin. 1972. Identification of T-2 toxin in moldy corn associated with a lethal toxicosis in dairy cattle. *Appl. Microbiol.* 24:682-690.
- Ishii, K., Y. Ando, and Y. Ueno. 1975. Toxicological approaches to the metabolites of fusaria. IX. Isolation of vomiting factor from moldy corn infected with *Fusarium* species. *Chem. Pharm. Bull.* 23:2162-2164.
- Joffe, A. Z. 1971. Alimentary toxic aleukia. p. 139-189. In S. Kadis, A. Ciegler, and S. J. Ajl (eds.) *Microbial toxins*, Vol. 7. Academic Press Inc., New York.
- Joffe, A. Z. 1974. Toxicity of *Fusarium poae* and *F. sporotrichioides* and its relation to alimentary toxic aleukia. p. 229-262. In I. F. H. Purchase (eds.) *Mycotoxins*. Elsevier Scientific Publishing Company, New York.
- Kellerman, T. S., W. F. O. Marasas, J. G. Pienaar, and T. W. Naude. 1972. A mycotoxicosis of *Equidae* caused by *Fusarium moniliforme* Sheldon. *Onderstepoort. J. Vet. Res.* 39:205-208.
- Korpinen, E. L. 1974. Mycotoxicological studies on *Stachybotrys alternans*. Ph. D. Thesis, Dept. Microbiol. Epizootol., Coll. Vet. Med., Helsinki, Finland.
- Kotsonis, F. N., E. B. Smalley, R. A. Ellison, and C. M. Gale. 1975. Feed refusal factors in pure cultures of *Fusarium roseum* 'gramini-arum'. *Appl. Microbiol.* 30:362-368.
- Mirocha, C. J., and S. Pathre. 1973. Identification of the toxic principle in a sample of poaeafusarin. *Appl. Microbiol.* 26:719-724.
- Mirocha, C. J., S. V. Pathre, B. Schauerhamer, and C. M. Christensen. 1976. Natural occurrence of *Fusarium* toxins in feedstuff. *Appl. Environ. Microbiol.* 32:553-556.
- Mitroiu, P., C. Grigore, V. Stoicea, P. Jivoian, and I. Cerni. 1973. Un ele aspecte ale stahihotriotoxicozei la taurine. *Rev. Zooteh. Med. Vet.* 13:66-73.
- Morooka, N., N. Uratsyji, T. Yoshizawa, and H. Yamamoto. 1972. Studies on the toxic substances in barley infected with *Fusarium* spp. *Jpn. J. Food Hyg.* 13:368-375.
- Naumov, N. A. 1916. Intoxicating bread. *Trudo Biuro po Mik. i Fitopat.* No. 12, p. 1-216, pls. I-VIII, Petrograd, 1916. Reviewed by M. Shapovalov. *Phytopathology* 7:384-386. 1917.
- Palyusik, M., K. Bamberger, and Z. Hagy. 1971. Experimental stachybotryotoxicosis of day-old and growing chickens with special regards to its diagnostic differentiation from fowl pox. *Mag. Allatorv. Lapja.* 26:304-306.
- Rodricks, J. V., and R. M. Eppley. 1974. *Stachybotrys* and stachybotryotoxicosis. p. 181-197. In I. F. H. Purchase (ed.) *Mycotoxins*. Elsevier Scientific Publishing Co., New York.
- Saito, M., and K. Ohtsubo. 1974. Trichothecene toxins of *Fusarium* species. p. 263-381. In I. F. H. Purchase (ed.) *Mycotoxins*. Elsevier Scientific Publishing Co., New York.
- Schumaier, G., H. M. DeVolt, N. C. Laffer, and R. D. Creck. 1963. Stachybotryotoxicosis of chicks. *Poult. Sci.* 42:70-74.
- Shadmi (Szmideberg), A., R. Volcani, and T. A. Nobel. 1974. The

- pathogenic effects on animals feed with mouldy hay or given its etheric fraction. Zentralbl. Veterinaermed. Reihe A. 21:544-552.
26. Snyder, W. C., and T. A. Toussoun. 1965. Current status of taxonomy in *Fusarium* species and their perfect stages. Phytopathology 55:833-837.
 27. Szabo, I., F. Rata, P. Aldasy, P. Szabo, and L. Gaal. 1970. Scaly and scabious skin affection and rhinitis (Stachybotryotoxicosis) in pig stocks. I. Clinical observations and aetiological investigations. Mag. Allatorv. Lapja 25:633-637.
 28. Szathmary, Cs. I., C. J. Mirocha, M. Palyusik, and S. V. Pathre. 1976. Identification of mycotoxins produced by species of *Fusarium* and *Stachybotrys* obtained from Eastern Europe. Appl. Environ. Microbiol. 32:579-584.
 29. Ueno, Y., K. Saito, and H. Tsunoda. 1970. Isolation of toxic principles from the culture filtrate of *Fusarium nivale*. p. 120. In Proc. 1st U.S.-Japan Conf. Toxic Microorganisms, Honolulu, Hawaii.
 30. Ueno, Y., I. Ueno, Y. Ditoi, H. Tsunoda, M. Enomoto, and K. Ohtsuba. 1971. Toxicological approaches to the metabolites of fusaria. III. Acute toxicity of fusaranon-x. Jpn. J. Exp. Med. 41:521-539.
 31. Vachev, V. L., X. X. Dyakov, P. Peichev, and B. Tabakov. 1970. Abortion caused by *Stachybotrys* toxin in swine. Vet. Slir. 67:7-10.
 32. Vesonder, R. F., A. Ciegler, and A. H. Jensen. 1973. Isolation of the emetic principle from *Fusarium*-infected corn. Appl. Microbiol. 26:1008-1010.
 33. Vesonder, R. F., A. Ciegler, A. H. Jensen, W. K. Rohwedder, and D. Weisleder. 1976. Co-identity of the refusal and emetic principle from *Fusarium*-infected corn. Appl. Environ. Microbiol. 31:280-285.
 34. Wyatt, R. D., J. A. Doerr, P. B. Hamilton, and H. R. Burmeister. 1975. Egg production, shell thickness, and other physiological parameters of laying hens affected by T-2 toxin. Appl. Microbiol. 29:641-645.
 35. Wyatt, R. D., P. B. Hamilton, and H. R. Burmeister. 1973. The effects of T-2 toxin in broiler chickens. Poult. Sci. 52:1853-1859.
 36. Wyatt, R. D., J. R. Harris, P. B. Hamilton, and H. R. Burmeister. 1972. Possible field outbreaks of fusariotoxicosis in avians. Avian Dis. 16:1123-1129.
 37. Wyatt, R. D., B. A. Weeks, P. B. Hamilton, and H. R. Burmeister. 1972. Severe oral lesions in chickens caused by ingestion of dietary fusariotoxin T-2. Appl. Microbiol. 24:251-257.
 38. Yagen, B. Toxins from a strain involved in alimentary toxic aleukia. Proc. Conf. on Mycotoxins in human and animal health. Univ. Md. Univ. Coll., Oct. 4-8, 1976, College Park, MD. (In press)
 39. Yagen, B., and A. Z. Joffe. 1976. Screening of toxic isolates of *Fusarium poae* and *Fusarium sporotrichiodes* involved in causing alimentary toxic aleukia. Appl. Environ. Microbiol. 32:423-427.