

Aflatoxin M in Milk..... A Review*

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Abstract. At present the dairy industries in Saudi Arabia are growing steadily and that emphasizes the need for giving special attention to the presence of aflatoxin M in dairy products. Recently the incidence of aflatoxin in animal feed ingredients has increased and that will increase the possibility of the occurrence of aflatoxin M in milk. However, aflatoxin M is a toxic (100% as AFB₁), Carcinogenic (33% of AFB₁), and mutagenic 3.3% of AFB₁) metabolite compound. This paper is a review of aflatoxin M in milk from lactating cows fed on aflatoxin-contaminated ration, where a linear relationship existed between the amount of AFB₁ ingested and the concentration of AFM₁ in milk.

Our interest was directed to review origin, excretion, biological effect, processing effects, and control of aflatoxin M in milk from lactating cows fed on aflatoxin-contaminated rations. The Food and Drug Administration (FDA) has established guidelines of 0.5 ppb for aflatoxin M in milk taking in the consideration that infants and young children are very likely to consume considerable quantities of milk and they are to be more susceptible to the carcinogenic effect of aflatoxin. The Saudi Arabian Standard Organization (SASO) is now in the process of establishing guidelines for aflatoxins in animal feeds to prevent occurrence of aflatoxin M in milk.

Introduction

Aflatoxins are considered at present to be one of the world's most dangerous contaminants in Foods and Feeds. Aflatoxins are a group of naturally occurring metabolites produced by the fungi *Aspergillus flavus* and *Aspergillus parasiticus* [1-2] where growth is favoured by a warm and humid climate [3]. The primary metabolites of *A.*

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flavus and *A. parasiticus*, are aflatoxins B1, B2, G1 and G2 which contaminate a variety of agricultural food and feed products [4, pp 13–54]. Investigation of food and feed stuffs began in 1960 when aflatoxins were discovered as the cause of the Turkey X disease that killed over 100,000 Turkeys in Britain [5]. Several investigators have found that aflatoxins are acutely toxic, carcinogenic, teratogenic, and mutagenic compounds [6-11]. Aflatoxin B1 is the most potent toxic and carcinogenic metabolite recognized and found naturally in large amounts in agricultural crops [11-13]. It is suspected that aflatoxins are primary source of human cancer in some population [14]. Aflatoxins in the food producing animals are of concern because they can be found in the food produced or obtained from those animals. It has been reported that the consumption of aflatoxin-contaminated feed by lactating animals results in the secretion of the toxic, carcinogenic and mutagenic aflatoxin M in the milk [15-17]. Feeding chickens ration containing aflatoxins cause growth retardation, and increase in liver/body weight ratio, biochemical alterations and pathological changes [18].

Aflatoxin M in milk can be determined either by the thin layer chromatography (TLC) or high pressure liquid chromatography (HPLC) separation. Several methods have been reported for the determination of the aflatoxin M in liquid milk [19-25] dry milk [26-28] and cheese [29,21].

Structures of aflatoxins

Aflatoxin M is the generic name of two compounds, designated aflatoxin M1 and M2. De Iongh *et al.* [30] showed that aflatoxin M is a blue-violet fluorescent compound which have an RF value about half that of aflatoxin B1 on Silica gel TLC using 3% methanol in chloroform as a solvent. Holzapfel *et al.* [31] reported that milk aflatoxin contained two fluorescent compounds subsequently designated as M1 and M2. Their structures were found to be hydroxylated derivatives of the aflatoxin B1 and B2 respectively (Fig. 1).

Biological effects of aflatoxin M₁

Aflatoxin B1 is the most potent toxic and carcinogenic metabolite recognized and found naturally in large amounts in agricultural crops [7,12-13]. It is suspected that aflatoxins are primary source of human cancer in some population [14]. The toxic dose of aflatoxin for humans is not established, but strong circumstantial evidence from Southeast Asia, India, and Africa, plus a suspect case in Germany, indicates that aflatoxins have been involved in human health, particularly among children [34-35].

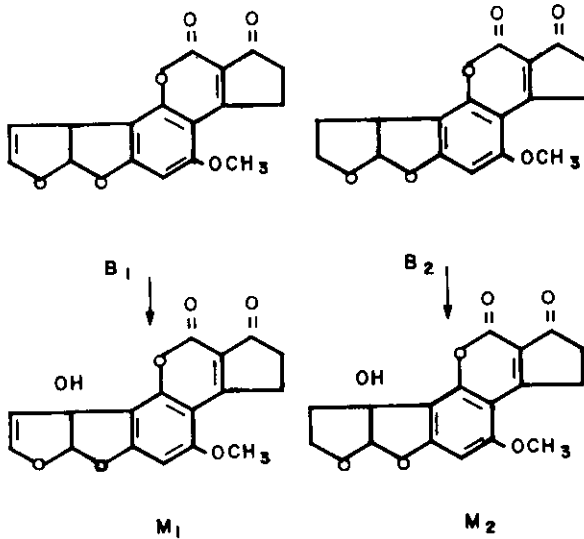


Fig. 1. Structures of aflatoxins obtained from Asao *et al.* [32] (AFB₁), Chang *et al.* [33] (AFB₂), Holzapfel and Steyn [31] (AFM₁ and AFM₂)

A reduction in milk production yield, loss of condition, and liver damage were observed in cows and heifer fed on rations containing 15 to 20% of the toxic groundnut meal for several months. Later estimates of aflatoxin M content were reported to be about 2 ppm with daily intakes of 3.2 mg B1 for dry cows and up to 12.7 mg B1 for milking cows [36]. In addition, severe illness and liver damage were observed in 2 years old fatten cattle given toxic groundnut meal for 3 months [37]. Calvet *et al.* [38] gave aflatoxin to lactating cows (1.5 mg/day) and studied the effect on the breast-fed calf. The nursing calves showed a reduction of growth, an increase in serum glutamic-oxaloacetic transaminase (GOT) and death of 3 of 5.

Allcroft and Carnaghan [1] observed liver damage in ducklings fed on an extract of freeze dried milk obtained from cows consuming aflatoxin contaminated ration. Purchase [39] reported that milk from cows exposed to aflatoxin B1 caused bile duct hyperplasia in one day old ducklings. Aflatoxin M1 acute toxicity in the day-old ducklings was found to be approximately the same as that of aflatoxin B1. Liver tumors were observed in two out of ten rats injected with AFM1 [40]. Pong and Wogan [41] injected male fisher rats with both synthetic and natural aflatoxin M1 and B1 (0.4–1.5 mg/kg) for 14 days. Their results indicated that the M1 acts through the same mechanism as aflatoxin B1 in causing toxicity and subcellular alterations. Syn-

thetic M1 and synthetic B1 were found to be of equal potency but both were less active than natural B1.

Sinnhuber *et al.* [42] and Canton *et al.* [43] studied the carcinogenicity of aflatoxin M1 in rainbow trout and their results revealed that the carcinogenic potency of aflatoxin M1 in rainbow trout was much lower than that of aflatoxin B1 (33% of AFB1). A similar result was reported by Wogan and Paglialunga [44], when they studied the carcinogenicity of synthetic aflatoxin M1 in rats (Fig. 2).

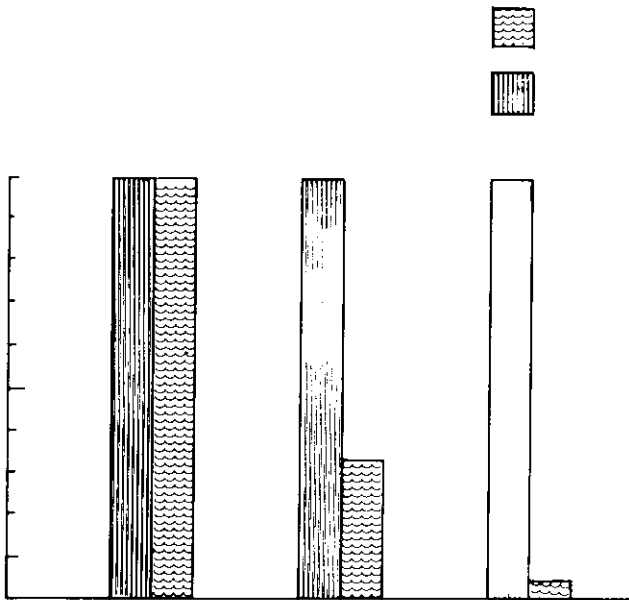


Fig. 2. Toxic, carcinogenic and mutagenic potency of milk aflatoxin (AFM₁) relative to aflatoxin B₁.
 a) From Pong and Wogan [41].
 b) From Sinnhuber *et al.*, [42] Canton *et al.* [43] and Wogan and Paglialunga [44].
 c) From Wong and Hsieh [45] and Uwalfo and Bababunmi [46].

Wong and Hsieh [45] determined the mutagenicity of the aflatoxins and various animal metabolites by using Ames tester Strain of *Salmonella typhimurium* (TA 98). Results indicated that aflatoxin M1 was found to have about 3.3% of the mutagenic potency of aflatoxin B1 (Fig. 2). Uwalfo and Bababunmi [46] studied the mutagenicity of aflatoxin M1 relative to that of aflatoxin B1 using five Ames tester strains of *Salmonella typhimurium* (TA 98, TA 100, TA 1535, TA 1537, and TA 1538). The

results obtained using the TA 98 Ames tester strain indicated that the mutagenicity of aflatoxin M1 relative to that of aflatoxin B1 is about 3%, which confirmed the results obtained by Wong and Hsieh [45] (Fig. 2).

Origin of aflatoxin M in milk

Aflatoxin M is a metabolite found in the milk of lactating cows consuming feed contaminated with aflatoxin. Feed ingredients were usually found contaminated with the naturally occurring aflatoxins, B1, B2, G1 and G2. Several studies indicated that aflatoxins had been detected in most animal feed ingredients such as corn, soybean, barley, cotton-seed and other common feed ingredients (Table 1).

Table 1. Some reports of aflatoxins – contaminated feed ingredients in different countries

Countries	Feed ingredients	References
Australia	Millet, Corn, Barley	[47-48]
Britain	Barley	[49]
Canada	Feeds and ingredients	[50]
Denmark	Imported peanut products	[51]
Finland	Imported peanut meal and copra	[52]
France	Rye, Barley, Soybean	[53]
Germany	Sugar Scrap, Linseed	[54]
India	Cottonseed, Safflower	[55,47]
Italy	Sesame (cake) Rapeseed, Sunflower seed (meal)	[47] [56]
Saudi Arabia	Poultry feeds, Imported corn	[57-58]
U.S.A.	Corn, Cottonseed, Wheat, Sorghum Oat, Linseed, Soybean	[59-62]

The incidence of aflatoxins in corn has increased [61] and this crop represents a major feed ingredient to all classes of livestock and poultry. That means, a special attention should be given to the incidence of aflatoxin in milk obtained from cows fed a ration containing corn.

Metabolism of aflatoxins by lactating cows

Aflatoxins in the ration of food-producing animals are of concern because they can be found in the food produced or obtained from those animals. Of practical concern to those in the dairy industries is aflatoxin M1. Several studies have reported that the consumption of aflatoxin-contaminated feed by lactating animals results in the secretion of the milk aflatoxin (AFM) in milk. Allcroft *et al.* [16], studied the

occurrence of aflatoxin in tissue, urine, and milk of sheep. Mixed aflatoxins, consisting of aflatoxin B₁, 36%; G₁, 52%; B₂, 3% and G₂ 2%, were dissolved in dimethylformamide and given to three yearling sheep (1 mg/kg). Urine and milk samples were also obtained and analyzed for the presence of aflatoxin and then animals were killed 2 hr. later, and the liver and kidney were removed so that analysis for aflatoxin could be performed by thin layer chromatography (TLC). Results showed that urine, liver and kidney extracts of all the sheep examined contained aflatoxins B₁ and G₁ and the "milk toxin" (AFM) while the milk extracts contained only the milk toxin.

Allcroft and Roberts [63] gave cows diets containing different concentrations of aflatoxin and followed the levels of aflatoxin M₁ (AFM₁) in the milk. The daily dose given to the cows ranged from 0.875–24.5 mg aflatoxin B₁. Results indicated the presence of AFM₁ in the milk and the quantity of this aflatoxin excreted in the milk was in direct proportion to the ingested aflatoxin B₁.

Masri *et al.* [15] gave 6 lactating cows a known amount of AFB₁ and monitored the level of AFM₁ in the milk. The source of AFB₁ was rice cultures which had been inoculated with *Aspergillus flavus*. The results showed a correlation between the dosage of AFB₁ and the concentration of AFM₁ in milk. Results also indicated that AFM₁ was excreted in the urine and feces of the cows in quantities comparable to that excreted in the milk.

Polan *et al.* [64] gave 4 lactating cows pure AFB₁ over a 14 day period. Aflatoxin B₁ was administered on a twice-daily basis at levels of 10, 50, 250 and 1,250 ppb of the concentrate. Obtained data indicated that, no AFM₁ could be detected in milk samples from cows receiving 10 ppb in the concentrate, although AFM₁ was detected in milk samples of cows given the two highest AFB₁ levels (250 and 1,250 ppb). They suggested that in order to ensure only negligible quantities of aflatoxin in milk, concentration of aflatoxin in the feed should not exceed 50 ppb. Only a trace amount AFM₁ (0.01 ppb) was found in milk samples of cows that received 50 ppb in the concentrate.

Patterson *et al.* [17] fed 6 lactating cows a ration contaminated with approximately 10 µg AFB/kg and followed the concentration of AFM₁ in milk samples which varied between 0.01 and 0.33 µg/L. Several studies [64,66] showed that aflatoxin M in the milk reached its maximum level between day 4 and day 6 after feeding the aflatoxin-contaminated ration (regardless of the aflatoxin dose) and then the level of AFM remained constant.

From the previous studies, it could be stated that a linear relationship exists between the amount of AFB₁ ingested and the concentration of AFM₁ in milk.

The other common aflatoxins in feed (B2 and G2) are converted similarly to compounds designated as M₂ and GM₂ in milk respectively [30,67].

The conversion ratio of AFB₁ in feed to AFM₁ in milk

Several studies have indicated that the levels of aflatoxin M₁ in milk are related to the levels of aflatoxin B₁ consumed in the feed of the lactating cows (Fig. 3). The lactating cows convert, in part, aflatoxin B₁ in the feed to a hydroxylated compound (AFM₁) (Table 2).

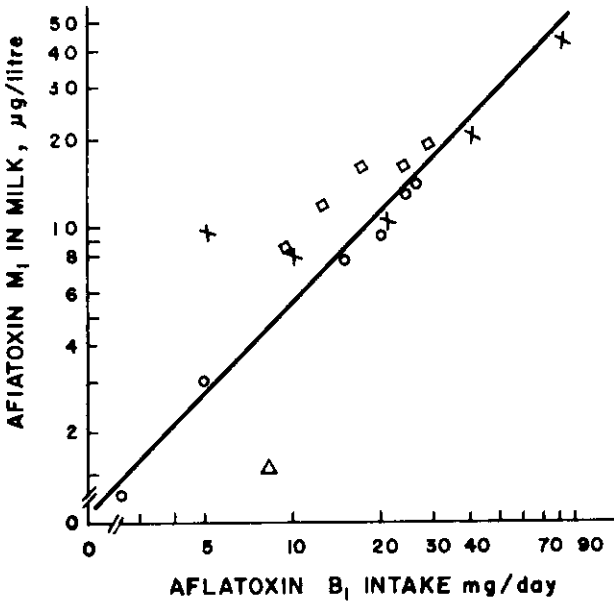


Fig. 3. Aflatoxin M₁ levels in milk from cows ingesting various amounts of aflatoxin B₁ obtained from Purchase [68]

Table 2. Conversion ratios of aflatoxin B₁ in feed to aflatoxin M₁ in milk

Conversion ratio (% M ₁ in milk/B ₁ in feed)	References	Conversion ratio (% M ₁ in milk/B ₁ in feed)	Reference
1.06	[66]	1.0	[69]
1.18	[66]	0.43–1.38	[65]
2.2	[17]	0.30 and 0.17	[64]
1.3	[15]	1.60	[70]

Masri *et al.* [15] fed lactating cows various levels of aflatoxin contaminated rice, and calculated the conversion ratios of aflatoxin B1 in the feed to aflatoxin M1 in milk. The results indicated that 1–3% of the ingested aflatoxin B1 is transmitted into the milk as aflatoxin M1. A similar result was reported by Van der Linde *et al.* [69] where they found less than 1.0% of the total amount of aflatoxin fed to lactating cows was converted to aflatoxin M. in milk.

Mckinney *et al.* [65] fed 4 lactating cows a ration containing aflatoxin contaminated cottonseed (5.82 mg AFB1/day). They found the conversion ratio of AFB1 to AFM1 to vary from 0.43 to 1.38% after a 72-hr. period during which maximum conversion of AFB1 to AFM1 was reached.

Ewaidh [66] gave lactating cows aflatoxin B1 – contaminated whole cottonseed (20.04 mg/day) and pure aflatoxin B1 (4.4 mg/day). Results indicated that, the conversion ratios of AFB1 to AFM1 were 1.06 and 1.18% respectively.

Polan *et al.* [64] gave 4 lactating cows pure AFB1 at a concentration of 250 and 1250 ppb on a twice-daily basis. Obtained results showed that the conversion of AFB1 to AFM1 was 0.30 and 0.17% respectively on day 4.

Price *et al.* [70] studied the conversion ratio of AFB1 in feed to AFM1 in milk by dairy cattle consuming naturally contaminated cottonseed (5, 31, 104, 157, 280 and 580 ppb AFB1) for 10 days. The average conversion ratio found was 1.6%.

Vander Linde *et al.* [69] indicated that aflatoxin M appears in cows milk within 2 days after ingestion of a ration contaminated with aflatoxin B1, also Allcroft and Roberts [63] reported that AFM can be detected in milk within 5 hr. after the administration of a dose of purified AFB1 by mouth.

This disappearance of aflatoxin M in the milk after withdrawal of aflatoxin contaminated feed or pure aflatoxin B1 have been studied by several investigators. Complete recovery of AFB1 in milk samples occurred 3–6 days after withdrawal of aflatoxin B1 from the ration (Table 3) [71,15,56,63,64,66,72].

Effect of milk processing on aflatoxin M levels

The first attempt to destroy aflatoxin in the milk was by heating treatments. Allcroft and Carnaghan [1] reported that toxicity of aflatoxin M in milk was not reduced by pasteurization (80°C for 45 sec. or 70°C for 30 min.) or by roller drying. Stoloff *et al.* [73] studied the effect of pasteurization treatment on aflatoxin M recov-

Table 3. Disappearance of aflatoxin M₁ from milk samples after withdrawal of aflatoxin B₁

Source of AFB ₁ in ration	Hours after withdrawal of AFB ₁ in ration							Reference
	Initial	24	48	72	96	120	142	
	Average ($\mu\text{g/L}$) AFM ₁ concentration in Milk							
Pure AFB ₁	4.40	1.10	0.17	0.06	0.00	0.00	0.00	[66]
Cotton seed	4.68	1.18	0.68	Trace	0.00	0.00	0.00	[65]
Cotton seed	1.80	1.55	1.29	0.65	0.41	0.05	0.00	[72]
Pure AFB ₁	0.85			0.00	0.00	0.00	0.00	[64]
or								
	0.38							

ery in raw and skim milk. Results showed that pasteurization of raw (63°C for 30 min.) and skim milk (77°C, 16 sec.) had no effect on aflatoxin M contents. Also, they reported that aflatoxin M₁ was recovered completely after storage of raw and pasteurized milk at 4°C for 17 days (Table 4). The same result was reported by Wiseman *et al.* [74] where they studied the effect of pasteurization of cream and skim milk made from naturally contaminated milk.

Applebaum and Marth [75] used hydrogen peroxide (H₂O₂), hydrogen peroxide plus riboflavin (Rib) and H₂O₂ plus lactoperoxidase (LPO) to inactivate aflatoxin M₁ in milk. They found that, maximum inactivation (98%) was obtained using 1% H₂O₂ plus 0.5 M Rib. (30°C, 30 min.) followed by heating at 63°C for 30 min. Also, the effect of H₂O₂ alone on stability of AFM₁ in milk was less compared with the effect of H₂O₂ plus lactoperoxidase (Table 4).

Recently, Yousef and Marth [76] irradiated the aflatoxin M₁ contaminated milk (1 ppb), which was held at 90°C for 10 min. and cooled to 20°C, with UV energy for 30 min. Aflatoxin M₁ content decreased by 56.2 and 53.9% in raw and pre-heated milk respectively.

In addition, attempts were made to separate aflatoxin M₁ from milk by manufacturing cheeses or yogurt. Several investigators [65,78,80-84] studied the presence of aflatoxin M₁ in various types of cheeses produced from aflatoxin M₁ contaminated milk. Obtained results indicated that aflatoxin M₁ has been found distributed between the curd and whey fractions obtained in making cheeses. The cheeses were found to have 2-7 times the aflatoxin M level of the milk samples from which it was made (Table 5). Butter, however will contain aflatoxin M₁ in proportion to the occluded whey component, which in turn can be expected to have an aflatoxin M level 0.5-0.7 times the level in the milk.

Table 4. Effect of hydrogen peroxide, pasteurization, refrigerated storage, ultraviolet energy and frozen storage on stability of aflatoxin M₁ in milk

Treatment	Aflatoxin level in milk	% inactivation of AFM ₁	References
Hydrogen peroxide 6% H ₂ O ₂ at 30°C for 30 min.	1 ppb (artificially contaminated 0.7–1.7 ppb (naturally cont.))	60% in the artificially contaminated raw milk, 83% in the artificially contaminated pasteurized skim milk, and 43% in the naturally conta- minated raw milk	[75]
Pasteurization 63°C for 30 min. (raw milk) 77°C for 16 min. (skim milk)	1.9 ppb skim milk	0% in raw milk and	[73]
Refrigerated storage 4°C for 17 days.	1.9 ppb skim milk	0% in raw and	[73]
Ultraviolet energy milk was held at 90°C for 10 min., cooled to 20°C and irradiated for 30 min.	1 ppb	53.9% in pre-heated milk 56.2% in raw milk	[76]
Frozen storage ice- cream and orange sherbet were made from naturally conta- minated milk and frozen for 8 months.	2.26–3.7 ppb (ice-cream) 0.96–2.32 ppb (sherbet)	0% in ice-cream and orange sherbet	[77]
Pasteurization cream and skim milk were made from naturally contaminated AFM ₁ and pasteurized at 46°C for 30 min.	1.1–2.6 ppb skim milk	0% in cream and	[74]
Pasteurization	1.57 ppb	0% in the raw milk	[78]
Sterilization	1.57 ppb	0% in the raw milk	[79]

It could be concluded that aflatoxin M₁ in milk is stable and unaffected by pasteurization, refrigeration, and processing of milk into cheese. In fact, it has been demonstrated that the aflatoxin M in cow's milk is associated with casein and remains with the milk portion when it is precipitated with rennin.

Aflatoxin M in commercial dairy products

Since there is a danger of the presence of aflatoxin M in the milk and of its toxic, carcinogenic and mutagenic effect, and because the infants and young children are

Table 5. Concentrations of AFM₁ in different kinds of cheese made from aflatoxin contaminated milk

Trial	AFM ₁ in milk ($\mu\text{g/L}$)	AFM ₁ in whey (μg)	AFM ₁ in Cheese ($\mu\text{g/L}$)	Increase of AFM ₁ in cheese	Reference
Parmesan cheese					
1	12.2	5.1	65.7	5.4 ×	[83]
2	11.8	4.8	63.7	5.4 ×	
3	5.7	1.5	35.4	6.2 ×	
Gouda cheese					
1	1.57	0.93	6.54	4.2 ×	[78]
Cheddar cheese					
1	0.70	0.30	3.07	4.32 ×	[84]
2	2.49	1.41	10.23	4.11 ×	
3	2.39	0.42	11.02	4.61 ×	
4	1.49	0.41	6.43	4.32 ×	
Mozzarella cheese					
1	1.5	<0.3	10.7	7.1 ×	[83]
2	1.8	<0.3	12.7	7.1 ×	
3	0.9	<0.3	6.5	7.2 ×	
Curd (Rennet precipitation)					
1	4.4	0.6	9.2	2.1 ×	[65]
2	4.4	0.5	9.3	2.1 ×	
3	4.4	0.6	9.2	2.1 ×	
Brick cheese (2 wk)					
1	3.7	4.6	9.6	2.6 ×	[82]
2	4.5	4.1	5.7	1.3 ×	
3	4.5	3.9	11.2	2.5 ×	

very likely to consume considerable quantities of milk, surveys of the incidence of aflatoxin M in milk have been undertaken. Allcroft and Carnaghan [1] were not able to detect contaminated milk from pooled milk samples in Great Britain. Many surveys were undertaken by several investigators [40,54,79,81,85-92] to find out if commercial dairy products in a number of countries are contaminated with aflatoxin M (Table 6).

Table 6. Survey of aflatoxin M₁ in dairy products from different countries

Dairy product and level of AFM ₁ (ppb)	Country	References
Raw and dried milk (0.02–0.2)	Belgium	(79)
Milk products (0)	France	[85]
Raw and market milk (0.04–0.25), Cheeses (0.02–0.23), Raw milk (0.05–0.54), Yogurt (0.05–0.74), Milk (0.02–2.0), Cheeses (0.1–1.30)	Germany	[54,87-89] [91]
Raw milk (0.005–0.091), heat treated milk (0.001–0.028)	Italy	[91]
Liquid and powdered milk (0.09–0.5)	Netherlands	[93]
Dried milk (0)	Norway	[92]
Retail milk (Trace –0.16)	S. Africa	[40]
Dried milk (less than 0.03), Liquid milk (less than 0.01)	U.K.	[94]
Cottage cheese (0.05–0.5), imported cheeses (0.1–1.0), whole milk (0) dried skim milk (0), evaporated milk (0)	U.S.A.	[86,90]

Control of aflatoxin M1 in milk

Initially the Food and Drug Administration (FDA) advice to control the occurrence of aflatoxin M in milk was to control the aflatoxin B1 contamination of feed. Emphasis has been placed on farm management practices such as using fungus-free seed, controlling insects and diseases, controlling irrigation practices, harvesting at maturity, and properly operating harvesting equipment to avoid damaging the crop [5]. The FDA established an action guideline of 20 ng/g (20 ppb) for total aflatoxins (B₁+B₂+G₁+G₂+) in all feeds or feed ingredients. Aflatoxin will not be detected in the milk from cows fed ration containing 20 ppb or less. The FDA gave extensive attention on aflatoxin in milk in 1977, when aflatoxin level rose above 20 ppb in two-thirds of the corn crop in U.S.A. and resulted in highly significant levels of Aflatoxin M1 in milk. The FDA established the action level of 0.5 ppb for the aflatoxin M1 contamination of fluid milk [95]. The reason for the significantly lower level allowed in milk was based on the consideration that infants and young children are very likely to consume considerable quantities of milk, and they are also likely to be more susceptible to the carcinogenic effect of aflatoxin.

Price *et al.* [72] found that ammoniation of aflatoxin-contaminated cottonseed at atmospheric pressure and ambient temperature was effective in reducing aflatoxin M1 level in milk obtained from cows fed on the ammoniated seeds. They treated the contaminated cottonseed with 1.5% ammonia and 10% water and packed it into a 3

m in diameter by 30 m long polyethylene bag for 21 days. Aflatoxin M₁ levels in milk from cows fed ammoniated (3.5 kg/day/cow) and non ammoniated cottonseed showed that ammoniation treatment was very effective in reducing the level of AFM₁ in milk below the FDA action level of 0.5 ppb (Fig. 4). Lee *et al.* [96] reported that, treatment of aflatoxin B₁ with ammonia resulted in the formation of a nonfluorescent phenol of molecular weight 286 (AFD₁). It is postulated that the new product arose from opening the Lactone ring of AFB₁ during ammoniation, formation of an ammonium salt of the resultant hydroxy acid, and loss of carbon dioxide from this B-Keto acid (Fig. 5). Although this reaction occurs in a test tube, in reality the situation is more complicated.

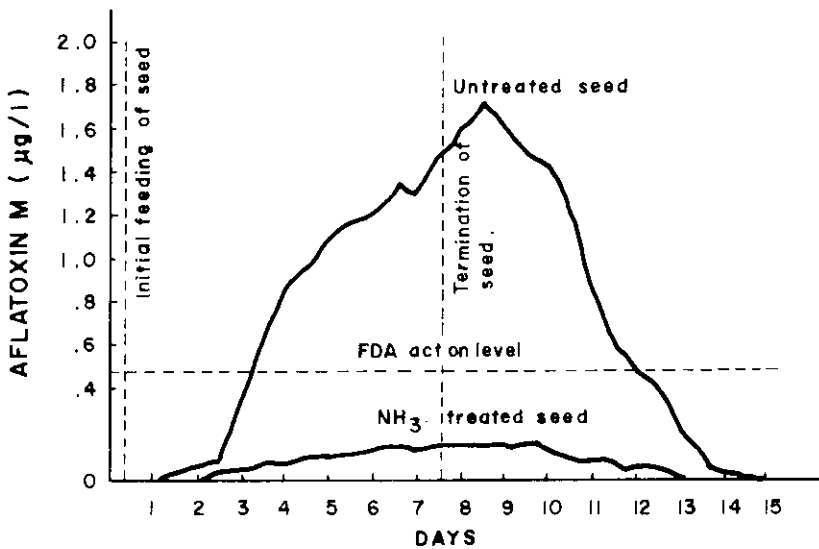


Fig. 4. Average aflatoxin M₁ levels at each milking of cows fed either ammonia-treated or non-treated, aflatoxin contaminated whole cottonseed. (after Prince *et al.* [72])

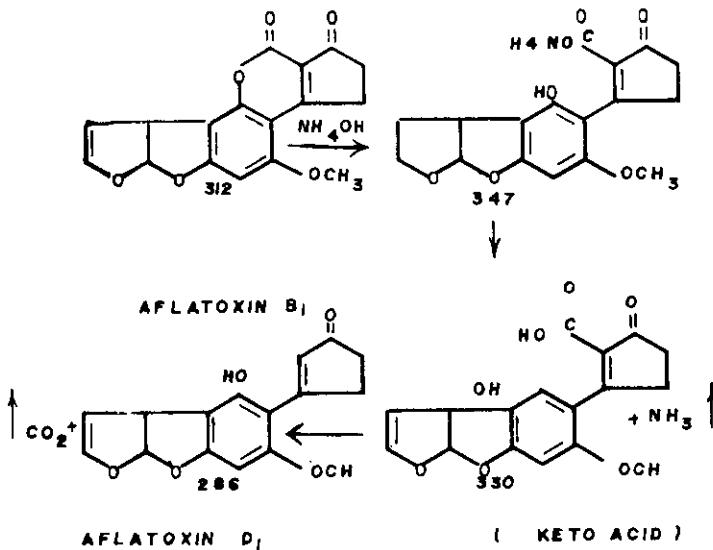


Fig. 5. Proposed scheme for formation of the major product of ammoniation of pure aflatoxin B₁. (after Lee *et al.* [96])

References

- [1] Allcroft, R. and Carnaghan, R.B.A. "Groundnut Toxicity; An Examination for Toxin in Human Food Products from Animals Fed Toxic Groundnut Meal." *Vet. Rec.*, **75** (1963), 259-263.
- [2] Sargeant, K., Sheridan, A., O'Kelly, J. and Carnaghan, R.B.A. "Toxicity Associated with Certain Samples of Groundnuts." *Nature*. **192** (1961), 1096-1097.
- [3] Schindler, A.F., Palmer, J.G. and Eisenberg, W.V. "Aflatoxin Production by *Aspergillus Flavus* as Related to Various Temperatures." *Appl. Microbiol.*, **15** (1967), 1006-1009.
- [4] Diener, U.L. and Davis, N.D. Aflatoxin formation by *Aspergillus flavus*, In: Goldblatt, L.A. (ed.) *Aflatoxin*. (ed.), NY: Academic Press, 1969.
- [5] Council for Agricultural Science and Technology (CAST) "Aflatoxin and Other Mycotoxins: An Agricultural Perspective." *Report No.80 ISSN 0194-4088* (1979).
- [6] Adamson, R.H., Correa, P. and Dalgard, D.W. "Brief Communication: Occurrence of Primary Liver Carcinoma in a Rhesus Monkey Fed Aflatoxin B₁." *J. Ann. Cancer Inst.* **50** (1973), 544-553.
- [7] Butler W.H. and Barnes, J.M. "Carcinogenic Action of Groundnut Meal Containing Aflatoxin in Rats." *Fd. Cosmet. Toxicol.*, **6** (1968), 134-141.
- [8] Keyl, A.C. and Booth, A.N. "Aflatoxin Effects in Liverstock." *J. Amer. Oil Chem. Soc.*, **48** (1971), 599-604.
- [9] Gopalan, C., Tulpule, P.G. and Krishnamurthy, D. "Induction of Hepatic Carcinoma with Aflatoxin in Rhesus Monkey." *Fd. Cosmet. Toxicol.*, **10** (1972), 519-521.
- [10] Lee, L.S., Stanley, J.B., Cucullu, A.F., Pons, W.A. and Goldblatt, L.A. "Ammoniation of Aflatoxin B₁; Isolation and identification of the Major Reaction Product." *JAOAC*, **57**, No. 3 (1974), 626-631.
- [11] Newberne, P.M. and Butler, W.H. "Acute and Chronic Effect of Aflatoxin on the Liver of Domestic and Laboratory Animals." *Cancer Res.*, **29** (1969), 236-250.

- [12] **Scott, P.M.** "Mycotoxins in Feeds and Ingredients and their Origin." *J. Food Prot.*, **41**, No. 5 (1978), 385-398.
- [13] **Wogan, G.N. and Newberne, P.M.** "Dose Response Characteristics of Aflatoxin B₁ Carcinogenesis in the Rat." *Cancer Res.*, **27** (1967), 2370-2376.
- [14] **Newberne, P.M. Rogers, H.E.** "Animal Model of Human Disease: Primary of Hepatocellular Carcinoma." *Am. J. Path.* **72** (1973), 137-146.
- [15] **Masri, M.S., Garcia, V.C. and Page, J.R.** "The Aflatoxin M Content in Milk from Cows Fed Known Amounts of Aflatoxin." *Vet. Rec.*, **84** (1969), 146-147.
- [16] **Allcroft, R. and Roberts, B.A.** "Toxic Groundnut Meal: The Relationship Between Aflatoxin B₁ Intake by Cows and Excretion of Aflatoxin M₁ in Milk." *Vet. Rec.*, **82** (1968), 116-118.
- [17] **Patterson, D.S.P., Glancy, E.M. and Roberts, B.A.** "The 'Carry Over' of Aflatoxin M₁ Into the Milk of Cows Fed Rations Containing a Low Concentration of Aflatoxin B₁." *Food Cosmet. Toxicol.*, **18** (1980), 35-37.
- [18] **Carnaghan, R.B.A., Levis, G., Paterson, D.S.P. and Allcroft, R.** "Biochemical and Pathological Aspects of Groundnut Poisoning in Chickens." *Pathol. Vet.*, **3** (1966), 601-615.
- [19] **Masri, M.S., Page, J.R. and Garcia, V.C.** "Analysis for Aflatoxin M in Milk." *J. Assoc. Off. Anal. Chem.*, **51** (1968), 594-600.
- [20] **Jacobson, W.C., Harmeyer, W.C. and Wiseman, H.G.** "Determination of Aflatoxin B₁ and M₁ in Milk." *J. Dairy Sci.*, **54** (1971), 21-24.
- [21] **Stubblefield, R.D.** "The Rapid Determination of Aflatoxin M₁ in Dairy Products." *J. Amer. Oil Chem. Soc.*, **56** (1979), 800-802.
- [22] **Yousef, A.E. and Marth, E.H.** "Rapid Reverse Phase Liquid Chromatographic Determination of Aflatoxin M in Milk." *J. Assoc. Off. Anal. Chem.*, **68**, No. 3 (1985), 462-465.
- [23] **Qian, G.S., Yasei, P. and Yang, G.C.** "Rapid Extraction and Detection of Aflatoxin M₁ in Cow's Milk by High-Performance Liquid Chromatography and Radioimmunoassay." *Analytical Chemistry*, **56** (1984), 2079-2080.
- [24] **Tyczkowska, K., Hutchins, J.E. and Hagler, W.M.** "Liquid Chromatographic Determination of Aflatoxin M₁ in Milk." *J. Assoc. Off. Anal. Chem.*, **67**, No. 4 (1984), 739-741.
- [25] **Takeda, N.** "Determination of Aflatoxin M₁ in Milk by Reversed-phase High Performance Liquid Chromatography." *J. Chromatogr.*, **288**, No. 2 (1984), 484-488.
- [26] **Purchase, I.F.H.** "Acute toxicity of Aflatoxin M₁ and M₂ in One Day Old Ducklings." *Food Cosmet. Toxicol.*, **5** (1967), 339-342.
- [27] **Carisano, A. and Torre, G.** "Sensitive Reversed-phase High Performance Liquid Chromatographic Determination of Aflatoxin M₁ in Dry Milk." *J. Chromatogr.*, **355** (1986), 340-344.
- [28] **Serralheino, M.L. and Quinta, M.L.** "Rapid Thin Layer Chromatographic Determination of Aflatoxin M₁ in Powdered Milk." *J. Assoc. Off. Anal. Chem.*, **68**, No. 5 (1985), 952-954.
- [29] **Shih, C.N. and Marth, E.H.** "A Procedure for Rapid Recovery of Aflatoxins from Cheese and Other Foods." *J. Milk Food Technol.*, **34** (1971), 119.
- [30] **De Jongh, H., Vles, R.O. and Van Pelt, J.G.** "Milk of Mammals Fed an Aflatoxin-Containing Diet." *Nature (London)*, **202** (1964), 466-467.
- [31] **Holzappel, C.W., Steyn, P.S. and Purchase, I.F.H.** "Isolation and Structure of Aflatoxins M₁ and M₂ in Milk." *Tetrahedron Letters*, **25** (1966), 2799-2803.
- [32] **Asao, T., Buchi, G., Abdel-Kader, M.M., Change, S.B., Wick, E.L. and Wogan, G.N.** "The Structures of Aflatoxins B₁ and G₁." *J. Am. Chem. Soc.*, **87** (1965), 882-886.
- [33] **Chang, S.B., Abdel-Kader, N.M., Wick, E.L. and Wogan, G.N.** "Aflatoxin B₂: Chemical Identity and Biological Activity." *Science*, **142** (1963), 1191-1192.
- [34] **Bourgeois, C.H., Shank, R.C., Grossman, R.A., Johnsen, D.O., Wooding, W.L. and Chandavimol, P.** "Acute Aflatoxin B₁ Toxicity in the Macaque and Its Similarities to Reye's Syndrome." *Lab. Invest.*, **24** (1971), 206-215.
- [35] **Ciegler, A.** "Mycotoxins: Occurrence, Chemistry Biological Activity." *Lloydia*, **38**, No. 1 (1975), 21-35.

- [36] **Goldblatt, L.A.** *Aflatoxin Scientific Background, Control and Implications*. New York: Academic Press, 1969.
- [37] **Clegg, F.G. and Bryson, H.** "An Outbreak of Poisoning in Store Cattle Attributed to Brazilian Groundnut Meal." *Vet. Rec.*, **74** (1962), 992-994.
- [38] **Calvet, H., Boudergues, R., Discacciati, E. and Cliche, M.** "Effects of Aflatoxin on the Milch Cow and on the Breast-fed Calf." *A report of the National Laboratory of Animal Breeding and Eterinary Research*, Dakar-Hann. (1966).
- [39] **Purchase, I.F.H.** "Aflatoxin Residues in Food of Animal Origin." *Food Cosmet. Toxicol.*, **10** (1972), 531-544.
- [40] **Purchase, I.F.H. and Vorster, L.J.** "Aflatoxin in Commercial Milk Samples" *S. Afr. med. J.* **42** (1968), 219.
- [41] **Pong, R.S. and Wogan, G.N.** "Toxity and Biochemical and Fine Structural Effects of Synthetic Aflatoxins M₁ and B₁ in Rat Liver." *J. National Cancer Institute.* **47** (1971), 585-590.
- [42] **Sinnhuber, R.O., Lee, D.J., Wales, J.M., Lander, M.K., Keyl, A.C.** "Aflatoxin M₁ a Potent Liver Carcinogen in Rainbow Trout." *Fed. Proc.*, **29** (1970), 568 (Abstr.).
- [43] **Canton, J.H., Kroes, R., Van Logten, M.J., Van Schothorst, M., Stavenutter, J.F.C. and Verhulsdonk, C.A.H.** "The Carcinogenicity of Aflatoxin M₁ in Rainbow Trout." *Food Cosmet. Toxicol.*, **13** (1975), 441-443.
- [44] **Wogan, G.N. and Paglialunga, S.** "Carcinogenicity of Synthetic Aflatoxin M₁ in Rats." *Food Cosmet. Toxicol.*, **12** (1974), 381-384.
- [45] **Wong, J.J. and Hsieh, D.P.** "Mutagenicity of Aflatoxins Related to Their Metabolism and Carcinogenic Potential." *Proc. Natl. Acad. Sci. USA*, **73** (1976), 2241-2244.
- [46] **Uwalfo, A.O. and Bababunmi, E.A.** "The Reduced Mutagenicity of Aflatoxin B₁ Due to Hydroxylation: Observations on Five *Salmonella typhimurium* Tester Strains." *Cancer*, **7** (1979), 221-225.
- [47] **Jones, B.D.** "Aflatoxin in Feedstuff Its Incidence, Significance and Control". *Proc. Conf. Anim. Feeds. Trop. Subtrop. Origin. Tropical Products Institute*, London, England. (1975), 273-290.
- [48] **Bryden, W.L., Rajion, M.L., Lloyd, A.B. and Cumming, R.B.** "Survey of Australian Feedstuffs for Toxicigen Strains of *Aspergillus Flavus* and for Aflatoxin." *Australian, Vet. J.* **51** (1975), 491-493.
- [49] **Shreeve B.J., Patterson, S.P. and Roberts, B.A.** "Investigation of Suspected Cases of Mycotoxicosis in Farm Animals in Britain." *Vet. Rec.*, **97** (1975), 275-278.
- [50] **Prior, M.G.** "Mycotoxin Determination on Animal Feedstuffs and Tissues in Western Canada." *Can. J. Comp. Med.* **40** (1976), 75-79.
- [51] **Krogh, P. and Hald, B.** "Forekomst of Aflatoksin i Importerede Jordnodprodukter (Occurrence of Aflatoxin in Imported Groundnut Products)." *Nord Veterinaermed*, **21** (1969), 398-407.
- [52] **Krogh, P., Hald, B. and Korpinen, E.L.** "Forekomst of Aflatoksin i Jordnodog Kokosprodukter Indfort tit Finland (Occurrence of Aflatoxin in Groundnut and Copra-Products Imported to Finland)." *Nord Veterinaermed*, **22** (1970), 584-589.
- [53] **Lafont, P. and Lafont, J.** "Contamination de Produits Cerealiers et d'Aliments due Betail par l'Aflatoxin." *Food. Cosmet. Toxicol.*, **8** (1970), 403-408.
- [54] **Kiermeier, F., Weiss, G., Behringer, G. and Miller, M.** "Uber das Vorkommen und den Gehalt von Aflatoxin M₁ in kasen des Handels." *Z. Lebensm. Unters. Forsch.*, **163** (1977a), 268-271.
- [55] **Vedanayagam, H.S., Indulkar, A.S. and Rao, S.R.** "Aflatoxin and *Aspergillus Flavus* Link in Ind. Cottonseed. *Indian J. Exp. Biol.*, **9** (1971), 410-411.
- [56] **Monaceli, R., Aiello, A., Di Muccio, A., Slavatore, G. and Datolo, G.** "Presenza di Aflattossina B₁ in Semi Oleosi Diversi dall Arachide." *Riv. Soc. Ital. Sci. Aliment.*, **5** (1976), 259-261.
- [57] **Ewaidah, E.H.** "Survey for Aflatoxins in Poultry Feed in Riyadh Region" (Under-Publication) (1987a).
- [58] **Ewaidah, E.H.** "Incidence of Aflatoxin in Imported Corn in Riyadh Region (S.A)," unpublished data. (1987b).
- [59] **Shotwell, O.L.** "Aflatoxin in Corn." *J. Am. Oil Chem. Soc.*, **54** (1977), 216-224.

- [60] **Schneider, S. de Leon, M., Garcia-prenedes, M. and Rolz, C.** "Determination of Aflatoxin and Aflatoxin-Producing Cultures in Recently Ginned Cotton-Seed in Central America." *J. Am. Oil Chem. Soc.* **49** (1972), 700-701.
- [61] **Shotwell, O.L., Hesselin, C.W., Burmeister, H.R., Kwolek, W.F., Shannon G.M. and Hall, H.H.** "Survey of Cereal Grains and Soybeans for the Presence of Aflatoxin. I. Wheat, Grain Sorghum and Oats." *Cereal Chem.*, **46** (1969), 446-454.
- [62] **Bean, G.A., Schillinger, J.A. and Klarman, W.L.** "Occurrence of Aflatoxins and Aflatoxin-Producing Strains of *Aspergillus spp.* in soybeans." *Appl. Microbiol.*, **24** (1972), 437-439.
- [63] **Allcroft, R. and Roberts, B.A.** "Toxic Groundnut Meal: The Relationship between Aflatoxin B₁ Intake by Cows and Excretion of Aflatoxin M₁ in Milk." *Vet. Rec.*, **82** (1968), 116-118.
- [64] **Polan, C.E., Hayes, J.R. and Cambell, T.C.** "Consumption and Fate of Aflatoxin B₁ by Lactating Cows." *J. Agric. Food Chem.*, **22** (1974), 635-638.
- [65] **Mckinney, J.D., Cavanagh, G.C., Bell, J.T., Hoversland, A.S., Nelson, D.M., Pearson, J. and Selkirk, R.J.** "Effects of Ammoniation on Aflatoxins in Rations fed Lactating Cows." *J. Am. Oil Chem. Soc.*, **50** (1973), 79-84.
- [66] **Ewaidah, E.H.** "Effect of Ammoniation Treatment of Aflatoxin B₁ on Mutagenicity and Levels of Aflatoxin M₁ in Milk." *Ph.D. dissertation.* The University of Arizona, Tucson. (1984).
- [67] **Patterson, D.S.P. and Roberts, B.A.** "The Formation of Aflatoxins B_{2a} and C_{3a} and Their Degradation Products during the *in Vitro* Detoxification of Aflatoxin by Liver of Certain Avian and Mammalian Species." *Food Cosmet. Toxicol.*, **8** (1970), 527-538.
- [68] **Purchase, I.F.H. and Steyn, M.** "Estimation of Aflatoxin M in Milk." *J. Assoc. Off. Anal. Chem.*, **50** (1967), 363-366.
- [69] **Van der Linde, J.A., Frens, A.M., Iongh H. de. and Vles, R.O.** "Inspection of Milk from Cows Fed Aflatoxin-Containing Groundnut Meal." *Tijdschr. Diergeneesk.*, **89** (1964), 1082-1088.
- [70] **Price, R.L., Paulson, J.H., Lough, O.G., Gingg, C. and Kurtz, A.G.** "Aflatoxin Conversion by Dairy Cattle Consuming Naturally-Contaminated Whole Cottonseed." *J. Food Prot.*, **48**, No. 1 (1985), 11-15, 20.
- [71] **Masri, M.S., Lundian, R.E., Page, J.R. and Garcia, V.C.** "Crystalline Aflatoxin M₁ from Urine and Milk." *Nature.* **215** (1967), 753-753.
- [73] **Price, R.L., Lough, O.G. and Brown, W.H.** "Ammoniation of the Whole Cotton-Seed at atmospheric Pressure and Ambient Temperature to Reduce Aflatoxin M₁ in Milk." *J. Food Prot.*, **45** (1982), 341-344.
- [74] **Stoloff, L., Trucksess, M., Hardin, N., Francis, O.J., Hayes, J.R., Polan, C.E. and Cambell, T.C.** "Stability of Aflatoxin in Milk." *J. Dairy Sci.*, **58**, No. 12 (1975), 1789-1792.
- [75] **Wiseman, D.W., Applebaum, R.S., Brackett, R.E. and Marth, E.H.** "Distribution and Resistance to Pasteurization of Aflatoxin M₁ in Naturally Contaminated Whole Milk, Cream and Skim Milk." *J. Food Protection.*, **46**, No. 6 (1983), 530-532.
- [76] **Applebaum, R.S. and Marth, E.H.** "Inactivation of Aflatoxin M₁ in Milk Using Hydrogen Peroxide and Hydrogen Peroxide Plus Riboflavin for Lactoperoxidase." *J. Food Prot.*, **45**, No. 6 (1982), 557-559.
- [77] **Yousef, A.E. and Marth, E.H.** "Use of Ultraviolet Energy to Degrade Aflatoxin M₁ in Raw or Heated Milk with and Without Added Peroxide." *J. Dairy Science*, **69**, No.9 (1986), 2243-2247.
- [78] **Wiseman, D.W. and Marth, E.H.** "Stability of Aflatoxin M₁ during Manufacture and Storage of Ice-cream and Sherbet." *Z. fuer Lebensmittel Untersuchung und Forschung*, **177**, No. 1 (1983), 22-24.
- [79] **Van Egmond, H.P., Paulsch, W.E., Veringa, H.A. and Schuller, P.L.** "The Effect of Processing on the Aflatoxin M₁ Content of Milk and Milk Products." *Arch. Inst. Pasteur.*, **54** (1977), 381-390.
- [80] **Van Pee, W., Van Brabant, J. and Joostens, J.** "Ladetection et le Dosage de 1 Aflatoxin M₁ Dans et le Lait en Pouder." *Rev. Agric.*, **30** (1977), 403-415.
- [81] **Stubblefield, R.D. and Shannon, G.M.** "Aflatoxin M₁: Analysis in Dairy Products and Distribution in Dairy Foods Made from Artificially Contaminated Milk." *J. Assoc. Off. Anal. Chem.*, **57** (1974), 847-851.

- [82] **Kiermeier, F.** and **Buchner, M.** "Verhalten Von Aflatoxin M₁ Während der Reifung and Lagerung Von Kase." *Z. Lebensm. Unters. Forsch.*, **164** (1977), 87-91.
- [83] **Brackett, R.E., Applebaum, R.S., Wiseman, D.W.** and **Marth, E.H.** "Fate of Aflatoxin M₁ in Brick and Limburger - like Cheese." *J. Food. Prot.*, **45** (1982), 553-556.
- [84] **Brackett, R.E.** and **Marth, E.H.** "Fate of Aflatoxin M₁ in Cheddar Cheese and in Process Cheese Spread." *J. Food Prot.*, **45**, No. 6 (1982a), 549-552.
- [85] **Brackett, R.E.** and **Marth, E.H.** "Fate of Aflatoxin in Parmesan and Mozzarella Cheese." *J. Food Prot.*, **45**, No. 7 (1982b), 597-600.
- [86] **Boutibonnes, P.** and **Jacquet, J.** "Sur la Frequnce de l'Aflatoxin et des *Aspergillus* dans les Aliments." *Compt. Rend. Soc., Biol.*, **163** (1969), 1119-1124.
- [87] **Brewington, C.R.** and **Wehrauch, J.L.** "Survey of Commercial Milk Samples for Aflatoxin M." *J. Dairy Sc.*, **53** (1970), 1509-1510.
- [88] **Kiermeier, F.** "Aflatoxin Residue in Fluid Milk." *Pure Appl. Chem.*, **35** (1973), 271-273.
- [89] **Polzhofer, K.** "Aflatoxin Bestimmung in Milch and Milchprodukten." *Z. Lebensm. Unters. Forsch.*, **163** (1977), 175-177.
- [90] **Kiermeier, F., Weiss, G., Behringer, G., Miller, M.** and **Ranfft, K.** "Vorkommen und Gehalt an Aflatoxin M₁ in Kasen des Handels." *Z. Lebensm. Unters. Forsch.*, **163** (1977b), 171-174.
- [91] **Rodricks, J.V.** and **Stoloff, L.** "Aflatoxin Residues from Contaminated Feed in Edible Tissues of Food Producing Animals." pp. 67-79. In: **Rodricks, J.V., Hesseltine, C.W.** and **Mehlman, M.A.** (eds.). *Mycotoxins in human and Animal Health*. Park Forest South, ILL: Pathodox Publishers, Inc., 1977.
- [92] **Visconti, A., Bottalico, A.** and **Solfrizzo, M.** "Aflatoxin M₁ in Milk, in Southern Italy." *Mycotoxin Research*, **1**, No. 2 (1985), 71-75.
- [93] **Yndestad, M.** and **Underdal, B.** "Aflatoksin i Nacringsmidler pa det Norske Market." *Nord. Veterinaermed*, **27** (1975), 42-48.
- [94] **Schuller, P.L., Verhulsetonk, C.A.H.** and **Paulsch, W.E.** "Aflatoxin M₁ in Liquid and Powdered Milk." *Zesz. Probl. Postep. Nauk Roln.*, **189** (1977), 255.
- [95] **Gilbert, J., Shepherd, M.J., Wallwork, M.A.** and **Knowles, M.E.** "A Survey of the Occurrence of Aflatoxin M₁ in UK - Produced Milk for the Period 1981-1983." *Food Additives and Contaminants* **1**, No. 1 (1984), 23-28.
- [96] **Food and Drug Administration** *Aflatoxin Contamination of Milk: Establishment of Action Level*. (4110-03). Fed. Register, 42: 234: 61630-61631. Washington, D.C.: U.S.Government Printing Office, 1977.
- [97] **Lee, L.S., Stanley, J.B., Cucullu, A.F., Pons, W.A.** and **Goldblatt, L.A.** "Ammoniation of Aflatoxin B₁: Isolation and Identification of the Major Reaction Product." *JAOCAC*, **57**, No. 3 (1974), 626-631.

أفلاتوكسين م في الحليب - عرض شامل

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ملخص البحث . تعتبر هذه الدراسة ملخصاً عن وجود الأفلاتوكسين من نوع م (AFM) في حليب الأبقار التي تغذت على علائق ملوثة بسموم الأفلاتوكسين إذ من المعروف وجود علاقة طردية بين كمية الأفلاتوكسين في العليقة وكميته في الحليب .

ونتيجة للتطور السريع في صناعة الألبان في المملكة العربية السعودية وانتشارها على نطاق واسع في الأونة الأخيرة كان من الضروري أن تغطي الدراسات حول احتمال وجود الأفلاتوكسين «م» في الحليب أو منتجاته باهتمام كبير. ومن المعروف أن الحليب يشكل الغذاء الرئيس للرضع والأطفال وكبار السن كما أن وجود الأفلاتوكسين «م» فيه يعتبر من المواد السامة التي تسبب الطفرات الوراثية والسرطانية .

وتتضمن هذه الدراسة عرضاً شاملاً لمصادر الأفلاتوكسين «م» وعملية إفرازه في الحليب وتأثيره البيولوجي على الحيوانات وتأثير التصنيع عليه .

وتنص تعليمات إدارة الأغذية والأدوية الأمريكية (FDA) على ألا يتجاوز تركيز الأفلاتوكسين في الحليب ٥٠ ميكروجرام /لتر. كما أن هيئة المواصفات والمقاييس السعودية بصدد إصدار مواصفات قياسية خاصة بالأفلاتوكسين في علائق الحيوانات تجنباً لظهوره في الحليب .