Analysis of Animal Feed Products Sold in Connecticut During 2023

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INTRODUCTION

The Animal Feed Regulatory Program Standards (AFRPS) were designed to institute a uniform framework for the establishment of animal feed monitoring programs at the state level. In Connecticut, The Agricultural Commodities Division of The Department of Agriculture (DoAg) is responsible for regulation and inspection of animal feeding products and pet foods. Products collected by Department of Agriculture agricultural commodity inspectors are delivered to the Department of Analytical Chemistry (DAC) at The Connecticut Agricultural Experiment Station (CAES) for analysis to ensure compliance with state and federal regulations. The laboratories in the Department of Analytical chemistry are ISO 17025:2017 accredited to perform percent crude fat, percent crude protein, pesticides, and Aflatoxin analysis in animal feeds. This accreditation ensures accurate results with appropriate quality control samples and record keeping.

Aflatoxins are carcinogenic toxins produced by the fungus *Aspergillus flavus*. *A. flavus* mold can grow on cereal grains and legumes such as corn or peanuts while in storage. In December 2020 there were 28 out of state fatalities linked to dog food contaminated with Aflatoxins (5). This event triggered an FDA recall of the associated animal feed batches. A similar event occurred in September of 2020, with a recall extending to animal feeds that used the same corn grain. There are four main Aflatoxins B1, B2, G1 and G2. Aflatoxin B1 is predominate and considered the most toxic. If ingested, B1 and B2 can be transformed into Aflatoxin metabolites M1 and M2 respectively, which can be transferred into milk. The toxins can also enter the human food chain both by direct consumption of the product, or through livestock that have eaten the contaminated product. Currently, the FDA has a set action level for combined Aflatoxins (B1+B2+G1+G2) ranging from 100-300 µg/kg for beef cattle, swine, or poultry fed animal feed containing corn, peanut, or cottonseed ingredients. A lower action level of 20 µg/kg is in place for animal feeds intended for dairy animals, immature animals, and pets (4).

Deoxynivalenol is a toxin produced by different Fusarium molds that can grow on wheat, corn, oats, barley, and other grains. The FDA has set advisory levels for Deoxynivalenol present in grains and grain by-products used to produce animal feeds. ranging from 5000 µg/kg to 10000 µg/kg for cattle, 10000 µg/kg for chickens, and 5000 µg/kg swine and other animals (6).

Fumonisin is a toxin produced by some strains of Fusarium that can grow on corn and sometimes wheat. The FDA has established guidelines for the total amount of Fumonisin (Fumonisin FB1 + FB2 + FB3) for corn intended for use in animal feeds. Limits range from 5000 µg/kg to 60000 µg/kg, depending

on the livestock type and age consuming the feed (7).

METHODS

In calendar year 2023, The Department of Analytical Chemistry at The Connecticut Agricultural Experiment Station analyzed 32 animal feed products for label guarantees, 31 products for the presence of Aflatoxins, 31 products for deoxynivalenol, 16 for Fumonisin, 56 samples for Macro Minerals, and 14 samples for pesticides analysis that were for sale in Connecticut. Animal feeds were collected by Department of Agriculture's Commodity Inspectors at manufacturing facilities, wholesale dealers, and retail locations. Samples were drawn from bulk storage/delivery containers, as well as from retail bags, boxes, and cans.

After delivery to The Connecticut Agricultural Experiment Station, the animal feed products were sub-sampled and prepared for analysis. The samples were analyzed for protein, fat, and fiber based on modified methods described in Official Methods of Analysis (2).

Aflatoxins were extracted using 80% acetonitrile in water, followed by solid phase extraction with cartridges specific for Aflatoxin B1, B2, G1, and G2. Analytes are then eluted using methanol. Aflatoxin analysis was performed using liquid chromatography with high resolution mass spectroscopy (LC-HRMS). The LC-HRMS detection limit for individual Aflatoxin compounds was 1 µg/kg.

Deoxynivalenol and Fumonisin were extracted using a modified QuEChERS (quick, easy, cheap, effective, rugged, and safe) method, using 10% formic acid in acetonitrile as the extraction solvent. Sample extracts were filtered then analyzed using a liquid chromatograph coupled to a high-resolution mass spectrometer (LC-HRMS).

Pesticide residues were extracted using modified QuEChERS method, using acetonitrile as the extraction solvent, followed by cleanup. Sample extracts were analyzed using both LC-HRMS and a gas chromatograph coupled to a tandem mass spectrometer (GC-MS/MS).

Samples analyzed for macro nutrients and toxic elements were prepared by closed vessel microwave acid digestion with analysis using Inductively Couples Plasma Optical Emission Spectroscopy (ICP-OES) and Inductively Coupled Plasma Mass Spectroscopy (ICP-MS).

All results are submitted to the Department of Agriculture for possible regulatory response.

RESULTS AND DISCUSSION

Label Guarantees

Table 1 shows the results for the analysis of crude protein, crude fat, and crude fiber. The label guaranteed nutrient value is denoted as (G) and the laboratory determined nutrient value is denoted as (R). Deficiencies and excesses of a guaranteed nutrient are expressed in red.
The number of samples deemed unsatisfactory in one or more macronutrients was 1 out of 32 samples processed (3.1%). In the unsatisfactory sample, mislabeling occurred for claimed % crude fat of 3% when analysis determined a value of 2.4% (Table 1). In one sample, analysis determined % crude fiber to be below quantitation level of 0.033%. In one sample, the guarantee is stated as N/A because it was not provided. (Table 1).
All protein samples are analyzed in duplicate. However, there are no duplicates for Fat and Fiber. The results for protein reflect the average of duplicates. If a protein or fat result is below the guarantee but passes this is

because the upper range of the method uncertainty result range was greater than the AAFCO minimum analytical variation (Table 1).

Aflatoxins, Deoxynivalenol, and Fumonisin

Animal feed products analyzed for Aflatoxins during 2023 are shown in Table 2. All 23 animal feed samples tested for Aflatoxins had a sum of Aflatoxins B1, B2, G1, and G2 that was below the limit of detection. Additionally, 21 of the feed samples tested for Fumonisin (Table 2) contained a sum of Fumonisin FB1, FB2, and FB3 that was below the limit of reporting. The reporting limit for FB1 was 1000 µg/kg, FB2 was 300 µg/kg, and for FB3 was µg/kg. However, of the 2 feed samples tested positive for the presence of Fumonisin FB1. Deoxynivalenol (Table 2), was detected in 18 feed samples above the reporting limit of 500 µg/kg but below a guidance level of 5000 µg/kg.

Pesticides

For the 2023 calendar year, 12 animal feed samples were analyzed for the presence of pesticide residues (Table 3). Only one of the tested samples contained pesticide residues above the reporting limit of 0.01 mg/kg. FED23-146 contained a non-violative amount of piperonyl butoxide and malathion (Table 3).

Elemental Analysis

A total of 56 samples of animal feed were analyzed for their elemental composition. Table 4 shows the results of element analysis. There were 3 deficiencies for macro mineral label claims. The first deficiency was in sample FED23-04 which claimed a minimum phosphorus level of 0.45 % when analysis determined a value of 0.36 % (Table 4). The second deficiency was in sample FED23-06 which claimed a maximum calcium level of 0.75 % when analysis determined a value of 1.35 % (Table 4). The third deficiency was in sample FED23-08 which claimed a maximum calcium level of 1.05 % when analysis determined a value of 1.48 % (Table 4).

Animal feed products are reported to be unsatisfactory based on guidelines established by the Association of American Feed Control Officials (AAFCO) (3). All samples were analyzed in duplicate for elemental analysis.

Table 1. A comparison between label guarantees (G) and analytical results (R) for animal feeds received from the Connecticut Department of Agriculture for the calendar year 2023. Values in red indicate results that did not pass.

Sample ID	Sample Description	Protein		Fat		Fiber	
		G	R	G	R	G	R
FED23-065	Animal Feed- Cat-Wet	8	10.1	5	8.2	2	0.12
FED23-067	Animal Feed- Cat-Wet	10	12.1	5	10.4	1.5	0.79
FED23-069	Animal Feed- Cat-Wet	12	13.7	2	3.3	0.2	< 0.033
FED23-071	Animal Feed- Cat-Wet	9	11.8	6.5	8	1	0.13
FED23-073	Animal Feed- Cat-Wet	8	10.2	3	**2.4**	1.5	0.33
FED23-075	Animal Feed- Cat-Wet	10	12.7	7	11.5	1	0.23

FED23-077	Animal Feed- Cat-Wet	10	10.6	5	11.7	1	0.36
FED23-079	Animal Feed- Cat-Wet	12	12.8	2	2.1	1.5	0.19
FED23-081	Animal Feed- Cat-Wet	8	9.8	3	5.3	1.5	0.44
FED23-083	Animal Feed- Cat-Wet	11	13.2	8	10	0.75	0.31
FED23-085	Animal Feed- Cat-Wet	7.5	8.8	6	6.8	1.5	0.39
FED23-087	Animal Feed- Cat-Wet	9	9.9	1.5	1.5	1.2	0.39
FED23-089	Animal Feed- Cat-Wet	13	12.2	0.2	0.9	1	0.21

Sample ID	Sample Description	Protein		Fat		Fiber	
		G	R	G	R	G	R
FED23-091	Animal Feed- Cat-Wet	7	10.5	3.5	5.2	4	1.1
FED23-093	Animal Feed- Cat-Wet	8	8.5	2	5.1	1.5	0.12
FED23-104P	Animal Feed-Cat -Dry	34	37.5	20	17.2	3.5	1.5
FED23-106P	Animal Feed-Cat -Dry	36	38.5	25	31.8	15	9.6
FED23-108P	Animal Feed-Cat -Dry	38	42.6	18	15.5	4.5	1.7
FED23-110P	Animal Feed-Cat -Dry	45	52.9	25	31.8	5	0.59
FED23-112P	Animal Feed-Cat -Dry	38	37.2	10	10.3	6	4.3
FED23-114P	Animal Feed-Cat -Dry	42	49.2	22	20.3	3.5	1.2
FED23-116P	Animal Feed-Cat -Dry	34	35.8	16	14.5	4	1.5
FED23-118P	Animal Feed-Cat -Dry	40	44.6	19	19.6	3.5	1.1
FED23-120P	Animal Feed-Cat -Dry	34	39.7	13	13.2	4	0.74
FED23-122P	Animal Feed-Cat -Dry	30	32.1	10	9.5	8	4
FED23-124P	Animal Feed-Cat -Dry	31	36.3	6	6.9	9	2.4
FED23-126P	Animal Feed-Cat -Dry	40	48.3	18	15.4	3	1.4
FED23-128P	Animal Feed-Cat -Dry	36	37.4	18	16.2	4	1.3
FED23-130P	Animal Feed-Cat -Dry	36	39.9	30	35.7	15	7.4
FED23-132P	Animal Feed-Cat -Dry	36	38.3	20	17.1	3.5	1.1
FED23-143	Animal Feed- Poultry	N/A	12.9	N/A	3.6	N/A	1.9
FED23-145	Animal Feed-Equine	12	12.5	2	2.5	28	27.1

Table 2. List of animal feeds tested for Aflatoxins B1, B2, G1, and G2; Deoxynivalenol; and Fumonisin FB1, FB2, and FB3 in the year 2023. Results in µg/kg.

Sample ID	Description	Total Aflatoxins	Deoxynivalenol	Total Fumonisin
FED23-059	Corn	<1	<500	<LOR
FED23-061	Soybean Meal	<1	1217	<LOR
FED23-062	Cracked Corn	<1	<500	<LOR
FED23-064	Soybean	<1	1987	<LOR
FED23-095	Soybean meal	<1	672	<LOR
FED23-096	Corn	<1	2106	<LOR
FED23-099	Soybean	<1	1914	<LOR
FED23-101	Cracked Corn	<1	2311	1554
FED23-133	Corn	<1	2345	<LOR
FED23-137	Soybean Meal	<1	1998	<LOR
FED23-138	Corn	<1	<500	<LOR
FED23-140	Soybean Meal	<1	2105	<LOR
FED23-141	Soybean Meal	<1	1727	<LOR
FED23-142	Soybean Meal	<1	1151	<LOR
FED23-148	Forage Extender	<1	<500	<LOR
FED23-150	Cracked Corn	<1	769	1342
FED23-151	Soybean	<1	1610	<LOR
FED23-152	Soybean	<1	1996	<LOR
FED23-154	Corn	<1	5518	<LOR
FED23-155	Soybean	<1	1963	<LOR
FED23-157	Corn	<1	<500	<LOR
FED23-159	Corn	<1	1128	<LOR
FED23-161	Corn	<1	5404	<LOR

Table 3. List of animal feeds tested for pesticides in the year 2023.

Sample ID	Description	Pesticide Found (mg/kg)
FED23-060	Corn	<0.01
FED23-063	Cracked Corn	<0.01
FED23-097	Corn	<0.01
FED23-100	Cracked Corn	<0.01
FED23-134	Corn	<0.01
FED23-139	Corn	<0.01
FED23-146	Forage Extender	Piperonyl Butoxide (0.036) Malathion (0.035)
FED23-149	Cracked Corn	<0.01
FED23-153	Corn	<0.01
FED23-156	Corn	<0.01
FED23-158	Corn	<0.01
FED23-160	Corn	<0.01

Table 4. Results of animal feeds tested for macro minerals in the year 2023. Values in red indicate results that did not pass.

Sample ID	Description	Moisture	Ca	K	Mg	Na	P	S	Cu	Fe	Zn	As	Cd	Hg	Pb
		%							mg/kg			ug/kg			
FED23-004	Animal Feed- Poultry	10.47	4.35	0.67	0.18	0.14	0.36	0.18	5	120.96	28.54	92.63	< 18.61	< 18.61	81.36
FED23-006	Animal Feed- Poultry	10.29	1.35	1.09	0.22	0.27	0.56	0.27	8	157.25	52.48	143.75	52.76	< 17.18	73.6
FED23-008	Animal Feed- Poultry	10.42	1.48	0.85	0.18	0.26	0.52	0.22	6	148.1	53.68	157.46	45.85	< 17.41	53.59
FED23-019	Animal Feed -Dog -Wet	84.89	0.25	0.15	0.02	0.11	0.22	0.1	4	90.31	95.18	8.86	5.59	< 3.10	5.6
FED23-021	Animal Feed -Dog -Wet	75.05	0.64	0.33	0.02	0.25	0.43	0.14	6	95.29	104.23	21.69	13.53	< 3.03	31.88
FED23-023	Animal Feed -Dog -Wet	77.6	0.46	0.21	0.04	0.13	0.35	0.12	4	55.1	47.68	17.26	16.73	< 3.04	13.15
FED23-025	Animal Feed -Dog -Wet	71.61	0.63	0.33	0.03	0.12	0.41	0.14	8	74.51	53.25	28.38	15.57	< 3.13	8.73
FED23-027	Animal Feed -Dog -Wet	78.18	0.48	0.24	0.02	0.24	0.28	0.14	7	65.14	57.32	22.13	13.27	< 2.99	30.54
FED23-029	Animal Feed -Dog -Wet	76.81	0.36	0.41	0.05	0.12	0.27	0.12	7	79.64	76.42	259.59	24.47	5.18	20.94
FED23-031	Animal Feed -Dog -Wet	79.37	0.5	0.18	0.02	0.22	0.34	0.13	5	62.24	61.3	3.4	5.77	< 3.12	7.72
FED23-033	Animal Feed -Dog -Wet	70.47	0.36	0.35	0.03	0.4	0.35	0.13	15	74.65	80.51	209.38	69.03	9.23	28.54
FED23-035	Animal Feed -Dog -Wet	74.53	0.69	0.31	0.03	0.21	0.44	0.15	4	49.83	67.04	6.43	5.86	< 3.09	10.61
FED23-037	Animal Feed -Dog -Wet	75.31	0.5	0.28	0.03	0.11	0.37	0.12	6	124.88	49.54	26.92	10.05	< 2.98	89.49
FED23-039	Animal Feed- Dog-Dry	4.73	2.12	0.87	0.12	0.55	1.05	0.38	7	106.96	86.32	122.84	102.17	< 20.92	65.62
FED23-041	Animal Feed- Dog-Dry	9.69	1.62	0.48	0.12	0.29	1.05	0.33	18	80.25	137.49	246.63	32.16	< 19.03	42.37
FED23-043	Animal Feed- Dog-Dry	5.21	1.94	0.77	0.11	0.55	1.06	0.47	11	241.71	188.87	141.1	40.15	< 17.94	256.03
FED23-045	Animal Feed- Dog-Dry	6.11	1.28	0.64	0.14	0.45	1.01	0.52	12	187.96	196.72	181.7	60.76	< 18.09	89.52
FED23-047	Animal Feed- Dog-Dry	5.41	1.85	0.68	0.14	0.36	1.23	0.49	15	187.46	121.32	29.32	80.24	< 17.10	94.48
FED23-049	Animal Feed- Dog-Dry	7.52	0.89	0.74	0.15	0.42	0.66	0.5	14	253.36	230.39	135.24	61.69	< 15.32	194.82
FED23-051	Animal Feed- Dog-Dry	5.14	0.79	0.88	0.2	0.22	1.02	0.4	39	242.8	71.59	57.24	120.9	< 17.99	93.79
FED23-053	Animal Feed- Dog-Dry	3.97	2.25	0.83	0.15	0.48	1.26	0.34	14	207.83	184.64	57.94	100.77	< 15.41	109.96
FED23-055	Animal Feed- Dog-Dry	7.1	1.56	0.79	0.11	0.4	1.09	0.33	16	168.5	184.07	21.79	34.49	< 17.04	125.81
FED23-057	Animal Feed- Dog-Dry	7.61	1.71	0.89	0.12	0.58	1.18	0.36	13	208.24	226.55	35.07	85.95	< 15.89	122.39

FED23-066	Animal Feed- Cat-Wet	72	0.58	0.25	0.04	0.44	0.38	0.17	4	109	106	161	21	< 3	16
FED23-068	Animal Feed- Cat-Wet	73	0.45	0.33	0.03	0.34	0.41	0.22	5	79	49	8	8	< 3	6
FED23-070	Animal Feed- Cat-Wet	81	0.16	0.22	0.02	0.1	0.15	0.19	4	47	27	1643	34	98	83
FED23-072	Animal Feed- Cat-Wet	77	0.8	0.26	0.03	0.13	0.49	0.16	6	69	62	7	5	< 3	11
FED23-074	Animal Feed- Cat-Wet	79	0.31	0.25	0.03	0.13	0.3	0.14	4	21	37	187	11	6	5
FED23-076	Animal Feed- Cat-Wet	74	0.35	0.39	0.02	0.12	0.2	0.17	12	113	105	6	11	< 4	18
FED23-078	Animal Feed- Cat-Wet	75	0.28	0.23	0.02	0.47	0.36	0.15	1	42	24	153	5	5	6
FED23-080	Animal Feed- Cat-Wet	81	0.29	0.18	0.02	0.09	0.24	0.16	4	68	43	878	18	38	32
FED23-082	Animal Feed- Cat-Wet	79	0.32	0.21	0.03	0.36	0.31	0.16	4	47	40	250	4	5	6
FED23-084	Animal Feed- Cat-Wet	74	0.8	0.24	0.03	0.12	0.49	0.17	5	69	51	45	32	4	5
FED23-086	Animal Feed- Cat-Wet	76	0.28	0.18	0.03	0.09	0.25	0.13	6	36	62	13	8	< 3	< 3
FED23-088	Animal Feed- Cat-Wet	84	0.23	0.22	0.02	0.08	0.19	0.13	7	22	59	11	< 4	< 4	25
FED23-090	Animal Feed- Cat-Wet	82	< 0.01	0.18	0.01	0.07	0.13	0.13	1	2	5	7	< 3	< 3	62
FED23-092	Animal Feed- Cat-Wet	76	0.28	0.2	0.02	0.1	0.25	0.17	4	74	70	25	16	< 3	26
FED23-094	Animal Feed- Cat-Wet	76	0.2	0.19	0.03	0.31	0.16	0.14	3	22	28	4	< 3	< 3	< 3
FED23-098	Animal Feed- Poultry	7	3.65	0.89	0.34	0.13	0.49	0.23	12	235	104	43	30	< 17	78
FED23-103MM	Animal Feed-Cat -Dry	5	1.53	0.81	0.12	0.42	1.1	0.5	24	274	342	236	54	< 17	213
FED23-105MM	Animal Feed-Cat -Dry	2	1.77	1.04	0.25	0.34	1.2	0.5	47	514	125	179	79	< 16	298
FED23-107MM	Animal Feed-Cat -Dry	6	2.61	0.78	0.14	0.48	1.71	0.48	18	134	166	62	28	< 17	26
FED23-109MM	Animal Feed-Cat -Dry	4	2.77	0.88	0.11	0.4	1.77	0.59	18	168	143	122	< 19	< 19	28
FED23-111MM	Animal Feed-Cat -Dry	5	1.8	1.14	0.18	0.34	1.27	0.47	12	269	132	1041	159	28	357
FED23-113MM	Animal Feed-Cat -Dry	6	2.92	1.36	0.15	0.45	1.77	0.62	26	264	168	275	54	< 15	364
FED23-115MM	Animal Feed-Cat -Dry	6	2.03	0.94	0.13	0.21	1.32	0.45	18	64	174	18	25	< 16	23
FED23-117MM	Animal Feed-Cat -Dry	5	1.63	0.85	0.16	0.79	1.31	0.61	20	234	131	1366	175	30	62
FED23-119MM	Animal Feed-Cat -Dry	7	1.68	0.83	0.1	0.52	1.18	0.49	12	152	100	186	36	< 16	91
FED23-121MM	Animal Feed-Cat -Dry	6	1.58	0.78	0.13	0.31	0.94	0.6	19	207	123	205	43	< 17	133
FED23-123MM	Animal Feed-Cat -Dry	8	0.91	0.69	0.07	0.43	0.67	0.74	13	162	197	40	56	< 17	56

FED23-125MM	Animal Feed-Cat -Dry	5	2.18	0.98	0.15	0.27	1.47	0.63	23	175	181	75	76	< 20	69
FED23-127MM	Animal Feed-Cat -Dry	7	1.8	0.76	0.13	0.55	1.39	0.44	19	110	237	1027	118	31	134
FED23-129MM	Animal Feed-Cat -Dry	1	1.21	0.71	0.21	0.16	1.06	0.36	5	89	49	23	22	< 19	42
FED23-131MM	Animal Feed-Cat -Dry	6	1.89	0.79	0.13	0.51	1.22	0.61	22	230	139	403	35	< 16	168
FED23-144	Animal Feed- Poultry	10	2.12	0.61	0.16	0.06	0.52	0.16	5	87	73	21	15	< 15	37
FED23-147	Animal Feed-Equine	10	1	1.25	0.34	< 0.01	0.39	0.14	7	359	54	33	126	< 17	69

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