APPLICATION NOTE

Food Safety - Lateral Flow

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Rapid Quantification of Total Aflatoxin Residues in Corn Using the AuroFlow AQ Afla Strip Test

Introduction

Aflatoxin is a group of mycotoxins originating from crops infected predominantly by *Aspergillus flavus* and/or *Aspergillus parasiticus* fungi. Aflatoxin contamination in food and feed threatens human and animal health, thereby requiring close monitoring by governmental agencies such as the USDA[1].

The AuroFlow™ AQ Afla Strip Test is a quantitative and rapid, six-minute lateral flow test kit designed to detect total aflatoxins (B1, B2, G1 and G2) for field or laboratory use. This kit utilizes an environmentally-friendly water-based extraction method and has received a USDA-FGIS certificate of conformance (#FGIS 2019-121) and approval for its high performance in corn samples[3]. This assay features a simple workflow and dynamic detection range from 2–300 ppb.

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Experimental

Materials and Methods

The AuroFlow AQ Afla Strip Test reagent kit was executed according to published USDA-FGIS Test Kit Instructions in conjunction with a portable and user-friendly QuickSTAR Horizon Strip Reader (PerkinElmer, CAT# FOOD-6006-01)[2, 4].

Reference materials produced from naturally contaminated corn and wheat were prepared at various aflatoxin contamination levels, certified using HPLC reference methods, and sourced from Trilogy Analytical Laboratory (Washington, MO, USA). Corn reference materials tested included 5.2 ± 0.8 ppb, 21 ± 2.9 ppb, 87.9 ± 11.9 ppb and 300 ± 18.6 ppb total aflatoxins. Certified reference standards to assess spiked commodity subtypes were sourced from Millipore Sigma.

Results and Discussion

Accuracy and Precision

84 total samples of corn at concentrations from 5–300 ppb were aliquoted into individual portions of 50 ± 0.2 g using an analytical balance (Sartorius MFR# ENTRIS224-1SUS) and subsequently tested by three operators (Table 1).

Table 1. Corn sample test results using the AuroFlow™ AQ Afla Strip Test. *Indicates testing with the dilution method for Highly Contaminated Samples.

Accuracy & Precision Evaluation – Corn (n=21 per conc.)					
CRM Expected Concentration (ppb)	Observed Range (ppb)		Mean (ppb)	Standard Deviation (ppb)	Relative Standard Deviation
	Lowest	Highest			
5.2	3.9	6.6	4.9	0.7	14%
21	16.1	23.4	19.8	1.8	9%
87.9	75.9	103.3	90.8	8	9%
87.9*	70.7	115.5	95.4	11.2	12%
300.0*	238.6	347.6	283.8	24.7	9%

Overall, the results obtained from the AuroFlow AQ Afla Strip Test compares well to the AOAC 994.08 official method data that was obtained from Trilogy Analytical Laboratory. All results meet the USDA Design Criteria and Test Performance Specifications for Quantitative Aflatoxin Test Kits[3].

Cross-Reactivity

An extraction was performed using the USDA-FGIS Test Kit Instructions for the AuroFlow AQ Afla Strip Test on an uncontaminated (ND, non-detect) corn sample. Mycotoxin reference standards were spiked into the ND liquid sample extract to evaluate a representative 5 and 20 ppb sample for each analyte. Table 2 outlines the resulting determinations for each analyte and their cross-reactivity in respect to aflatoxin B1. Note that approximately 90% of naturally-occurring total aflatoxin is comprised of aflatoxin B1, followed by aflatoxin B2 at 5-10% incidence.

Table 2. Cross-reactivity profile of the AuroFlow™ AQ Afla Strip Test.

% Cross-Reactivity (n=10)		
Aflatoxin	B1	100%
	B2	37%
	G1	69%
	G2	23%

Sample Variability

Representative samples for each commodity subtype were ground in accordance to the suggested USDA-FGIS guidelines for sampling and processing. The resulting samples were then analyzed by Trilogy Analytical Laboratory (Washington, MO, USA) for confirmation of a natural ND level of contamination before proceeding with any further testing. A single extraction was then performed for each commodity subtype in accordance with The AuroFlow AQ Afla Strip Test and the associated USDA-FGIS Test Kit Instructions. An aflatoxin B1 reference standard was then spiked into the ND liquid sample extract in order to simulate a representative 5.0 and 20 ppb sample for each commodity subtype. The results are as shown in Table 3.

Table 3. Sample variability assessment of common commodity subtypes for corn using the AuroFlow AQ Afla Strip Test.

Commodity Sample Variability - Corn					
Contamination Level (Spike)	Commodity Subtype	AuroFlow AQ Afla Strip Test, Reported Values (n=10)			
		Mean (ppb)	Standard Deviation (ppb)	Relative Standard Deviation	% Match*
N/A	Flour	2.1	0.3	14%	N/A
	Corn Meal	2	0.3	15%	95%
	Grits	2	0.1	5%	95%
	Polenta	0.7	0.4	54%	33%
	Deer Corn	1.4	0.9	64%	67%
5.0 ppb	Flour	5.6	0.6	11%	N/A
	Corn Meal	4.7	1.2	26%	84%
	Grits	2.9	0.4	14%	52%
	Polenta	4	0.5	13%	71%
	Deer Corn	4.3	0.6	14%	77%
20 ppb	Flour	27.7	1.1	4%	N/A
	Corn Meal	26.1	1.7	7%	94%
	Grits	21.4	0.8	4%	77%
	Polenta	23.6	0.4	2%	85%
	Deer Corn	24.2	0.8	3%	87%

* % Match was computed against the corn 'Flour' with respect to each additional commodity subtype.

Conclusion

In summary, the AuroFlow AQ Afla Strip Test is an accurate, precise, and stable product for detection of total aflatoxin. The kit is easy to use and capable of determining total aflatoxin concentrations in corn and corn derivatives using an aqueous extraction method. Conformance of this product to USDA-FGIS aflatoxin test kit criteria in both internal and external testing

(USDA FGIS 2019-121) provides consumers with an ideal solution in rapid aflatoxin test applications.

References

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2. USDA Agricultural Marketing Service - Federal Grain Inspection Service. FGIS Performance Verified & Approved Mycotoxin Test Kits – Effective 06/13/2019. Retrieved from https://www.ams.usda.gov/sites/default/files/media/FGISApprovedMycotoxinRapidTestKits.pdf (2019, June 14).
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