

Introduction:

Aflatoxins are produced as secondary metabolites by the fungus *Aspergillus flavus* and *A. parasiticus* on variety of food products. They are potent toxic, carcinogenic, mutagenic, immunosuppressive and have been associated with various diseases, such as aflatoxicosis, in livestock, domestic animals and human throughout the world. There are four major aflatoxins, B1, B2, G1, G2 plus two additional metabolic products, M1 and M2, that are of significance as direct contaminants of foods and feeds. Aflatoxin B1 (AFB1) is normally predominant in amount in cultures as well as in food products. Food products contaminated with aflatoxins include cereal, oilseeds, spices, tree nuts and milk. Regarding the amount of aflatoxins allowable in human and animal foodstuffs, many countries have attempted to limit exposure to aflatoxins by imposing regulatory limits on commodities intended for use as food and feed. Thus it is very important to develop an rapid and accurate detection tool for detecting the presence of aflatoxin in commodities.

INTENDED USE:

Aflatoxin B1 Test is a qualitative immunochromatographic assay for the detection of aflatoxin B1 in cereals, maize, groundnuts, spices, feeds and other commodities.

PRINCIPLE:

The basis of the immunochromatographic assay in test cassette is an competitive antigen-antibody reaction. The aflatoxin molecules in the samples and AFB1-BSA fixed on the membrane(T-zone) will compete to combine the red complex of specific immunogold aflatoxin B1 antibodies. The results are read visually by observing the development of colored bands. The control band (control line) is not influenced by aflatoxin in the sample and should be present in all cases in order to prove the test card is valid. The test band (test line) is clearly visible in the absence of aflatoxin in the sample and if aflatoxin is present in concentrations of 5 ppb* and higher the band is invisible or very fade.

MATERIALS PROVIDED:

- A-40 aluminum foil pouches each containing Af B1 test card.
- B-40 tubes containing buffer A.
- C- 40 micro tubes containing buffer B.
- D- 40 droppers
- E- One instruction for use.

MATERIALS AND INSTRUMENTS NOT PROVIDED:

- A-small Kitchen grinder.
- B- Small Kitchen balance (accuracy of 1 grams).

PRECAUTIONS:

- All specimens should be handled as being potentially poisonous.
- Do not open or remove test kit from their individually sealed pouches until immediately before their use
- Do not reuse test kit.
- All reagents must be at room temperature before running the assay.
- Do not use reagents beyond the stated expiration date marked on the package label.

The components in this kit have been quality control tested as standard batch unit.
-Do not mix components from different lot numbers.

STORAGE INSTRUCTIONS:

The kit can be stored at room temperature (2-30°C) or refrigerated. The test kit is stable through the expiration date marked on the package label. **DO NOT FREEZE**. Do not store the test kit in direct sun light .

PROCEDURE OF THE TEST:

1-Please carefully read the instruction prior to testing. Bring all the reagents to room temperature before use.

2-Remove the test card from the foil pouch. Place it horizontally on the desk. Please use it soon in one hour.

EXTRACTION PROCEDURE:

1. Cereals:

A-Grind a small spoonful or 1 gram of sample to the particle size of fine instant coffee .

B-Transfer the powder into Bottle A ,shake for 1 min. and let to stand for 3 min at RT .

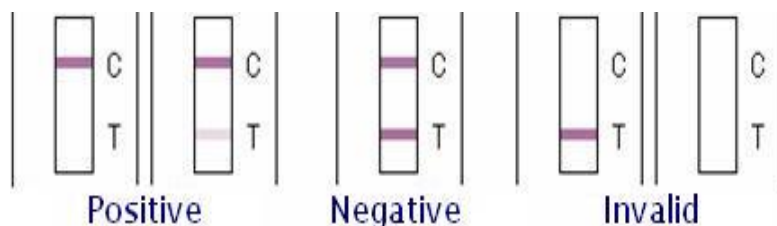
C-Transfer 4 drops of supernatant to tube B (use the dropper provided or otherwise use the dropper provided on the top of container), shake for 1 min.

D- Add 3 to 4 drops of resulting solution to the well denoted by "S" on the cassette (use the dropper) .

E- Interpret test results at about 8 minutes. Do not interpret after 15 minutes.

INTERPRETATION OF RESULTS:

The presence of only one **clear** band in zone "C" indicates a positive result. That means the level of aflatoxin B1 is at or over 5ppb in the sample. If one very **weak** band appears in zone "T", the result is also considered positive. The presence of two clear color bands ("T" and "C") within the result window indicates a negative result and tells that the concentrations of aflatoxin B1 is no more than 5ppb in the sample. If the control band is not visible in zone "C", the result is considered invalid (please see below) .



LIMITATION

The Rojan Aflatoxin Test is a useful tool offering a rapid and accurate testing in practical detection. The result is usually considered as a primary estimation. Other testing methods should be followed to give a comprehensive estimation if necessary.

Rojan Azma Research and mfg Co .No 40 , Golestan No 4, Baharestan Industrial Estate ,5thkm Karaj –Qazwin High Way Alborz- Iran . Phone :009826-34760503 Fax: 009826-34760610 Email : info@rojanazma.ir web site : www.rojanazma.ir