

EZtest™ Afla Total Strip for Quantitative Detection of Total Aflatoxin (Peanuts)

(Aflatoxin is a class I carcinogenic substance)

EZtest™ Strip-103P MANUAL

(v. 1.00)





INTRODUCTION

Aflatoxins are toxic and carcinogenic. They are metabolites of the fungi $Aspergillus\ flavus$ and $Aspergillus\ parasiticus$. There are four principle types of aflatoxin: B_1 , B_2 , G_1 and G_2 , which are named for their respective innate fluorescent properties. Aflatoxin B_1 is the most frequently encountered of the group and the most toxic. Aflatoxin B_1 is the most potent liver cancer-forming chemical known. It is a product of a mold called $Aspergillus\ flavus$, which is found in food that has been stored in a hot and humid environment. This mold is found in such foods as peanuts, rice, soybeans, corn, and wheat.

1 INTENDED USE

EZtest[™] Afla Total strip is lateral flow immunochromatographic assay based on a competitive immunoassay format. With EZtest[™] Stripreader, it measures 2.5ppb-25ppb of total aflatoxin B1, B2, G1 and G2 in peanut.

2 KIT CONTENTS

- 1) EZtest[™] Afla Total strips (25/pk)
- 2) Dilution buffer (10ml)
- 3) 1.5ml Strip Test Vial (50)
- 4) 1ml Pipette Tip (25)
- 5) 200µl Pipette Tip (50)
- 6) Calibration RFID card
- 7) Manual

Materials required but not provided:

- 1) 200 and 1000µl precision micropipette
- 2) EZtest[™] Stripreader (with ZERO card)
- 3) Vortex mixer
- 4) Multi-purpose rotary mixer
- 5) Timer



- 6) Incubator (37°C)
- 7) Weigh scale
- 8) Distilled water or pure water
- 9) Extraction solution: Methanol+Water(70+30)
- 10) Funnel
- 11) Filter paper
- 12) 50mL Extraction tube

3 PREPARATION

- 1) Store test kits at $4\sim8^{\circ}$ C (39 $\sim42^{\circ}$ F) when not in use, and do not use beyond the expiration date.
- 2) Keep from moisture, and do not use the damaged and damp kits.
- 3) All reagents must be at room temperature before assay is running.
- 4) Do not re-use test strips.
- 5) Avoid swallowing the desiccant.

4 SAMPLE PREPARATION

- 1) Grind samples of peanut to pass a 20 mesh sieve and thoroughly mix prior to sub-sampling. Samples not being immediately analyzed should be stored refrigerated.
- 2) Weigh 5.0 g ground sample and transfer to a 50mL Extraction tube.
- 3) Add 25 mL of 70% Methanol/water to the Extraction tube.
- 4) Mix it with rotary mixer for 5 minutes.
- 5) Filter 1-5 mL through a paper filter and transfer 100µl of extract to a 1.5ml Strip Test Vial.
- 6) Dilute 100μl of extract with 400μl of dilution buffer, and mix thoroughly with vortex mixer for 10 seconds.

5 TEST PROCEDURE

- 1) Please read the manual carefully before assay is running.
- 2) All reagents and kit components must be at room temperature $18-30^{\circ}$ C (64-86°F) before use.
- 3) Calibrate zero for EZtest™ Stripreader with ZERO card.
- 4) Calibrate EZtest™ Stripreader with RFID card.
- 5) Remove strip sealer, and take out the test strip on a flat surface (such as a countertop).



- 6) Add 100µl of the sample extracts to the circular opening of strip.
- 7) Put the test strip in Incubator (37 $^{\circ}$ C) to develop color for 10 minutes.
- 8) Put the test strip into the EZtest™ Stripreader, and the result can be read immediately.

6 INTERPRETATIONS OF THE RESULTS

- 1) Test range: 2.5ppb-25ppb.
- 2) If there is no line in control zone, the test is invalid and the sample should be re-tested by using a valid test strip.

7 Contact Information

UC BIODEVICES CORP.

3652 Edison Way Fremont, CA 94538

USA

Tel: 1-510-730-2598 Fax: 1-510-795-1795 www.ucbiodevices.com

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