

NEOVENTURES BIOTECHNOLOGY INC.

Afla-Sense® Detection for Corn and Peanuts 1ppb-200ppb

Product # 20540

Intended Use

The Afla-Sense® System for corn and peanuts is a fluorescent system used for the quantification of total aflatoxins in grain samples.

Aflatoxin

Assay Principles

The Afla-Sense® System is a fluorescent based detection system. Total aflatoxin is extracted from a ground sample with 80% methanol. The diluted extracts are purified through the Afla-Sense® Affinity Column. The elutant is mixed with a fluorescence enhancer and the sample is placed in a microplate well and read in a fluorometer at an excitation of 370 nm and an emission of 440nm. The intensity of the fluorescence signal is directly proportional to the concentration of total aflatoxin in the sample. The relative fluorescence units (RFUs) are compared to the RFUs of the standards and an interpretive result is determined.

Precautions

1. Adhere to protocols exactly stated. Alteration of the protocol may give inaccurate results.
2. Methanol is flammable. Caution must be taken in its use and storage.
3. The Afla-Sense® Buffer B contains Tris which is an irritant. Avoid contact with skin or eyes.
4. Consider all materials, containers and devices that are exposed to the sample or standards to be contaminated with toxin. Wear protective gloves and safety glasses when using the kit.
5. Dispose of all materials, containers and devices appropriately after use.

Setting up the BMG FLUOstar Omega

Changing the filter positions:

- 1) Under the “setup” tab, select “filters”
- 2) Type in “BMG” for the password. It will automatically allow you to edit the filter positions.
- 3) Type in the value of the corresponding filter in the correct position.
Example, position 2 excitation 380.
- 4) Select “OK”

Adding a microplate:

To add the Corning 96 well microplate settings:

- 1) Under the “setup” tab, select “Microplates...”
- 2) Select “New”
- 3) Name your plate Corning 96
- 4) Select plate format: 96
- 5) Input the following numbers in the following area for XY dimension:
Length: 127.8 Width: 85.50
Corner well X: 14.30 Corner well Y: 11.2
Well shape: round Well Diameter 6.86
- 6) Input the following numbers in the following area for Z dimensions:
Plate height H: 14.20
Well depth W: 10.67
Border Height B: 6.09
Distance between plate and well bottom D: 2.26
Stack height S: 12.97

Sample preparation

Refer to the product insert for AflaSense® Ultrafast columns.

Detection

1. Prepare the 3 standards, 0, 12, 24 ppb by adding 500* μ L of each standard to a new 1.5mL tube.
2. Prepare detection enhancement solution by diluting the AflaSense® Detector solution by 1/25 with H₂O. Prepare fresh solution everyday. 20 μ L of diluted solution is required per sample.



3. Add 20* uL of diluted detector solution to the standards and to the samples. Mix well.
4. Incubate the samples and standards for 10minutes at 70 degrees Celcius. Cover the tubes with aluminum foil to prevent photo-degradation of the sample.
5. Cool the samples and standards to room temperature by placing samples in -20 degrees C for 3 minutes or placing the samples on ice for 1 minute.
6. Mix the samples well and load 300 uL onto an approved microplate.
7. Read the samples at the following settings:
 Reading type: Fluorescence intensity endpoint
 Microplate: Corning 96 plate
 Position delay: 0.2
 Measurement start time: 0
 Optic: Top Optic
 No. of multichromatics: 1
 Excitation 370
 Emission 440
 Gain 1250
 In layout, select the wells to be read for the experiment.

*To maximize use of the kit when running less than 4 samples at once, use 300ul of elution with 12uL of diluted detector solution. A minimum of 3 samples may be used.

Interpretation of data

Refer to the Excel spreadsheet provided by NeoVentures Biotechnology Inc. Follow the instruction given on the spreadsheet. Please contact us if the spreadsheet has not been provided upon receipt of the kit or if further instruction is required.

Performance Characteristics

Limit of detection (LOD) : 0.5 ppb
 Range of quantitation: 1-200ppb

Materials supplied with kit (25 samples)

- 1 vial of AFLA Sense Detector
- 1 vials of 4mL of each aflatoxin standard (0, 12 and 24 ppb) 3 vials total.
- AFLA-Sense UltraFast columns (25 columns)

Materials required but not provided with kit

- Single channel pipettes capable of pipetting 20µL, 300µL, and 500µL volumes with tips
- Black low-fluorescent microplate (Corning® or comparable)
- Approved fluorometer
- Microfuge tubes
- Heating block/water bath capable of heating to 70 degrees Celcius.
- Ice or -20 degree freezer.

Warranty

The user assumes all risk in using NeoVentures Biotechnology Inc. products and services. NeoVentures Biotechnology Inc. will, at its option, repair or replace any product, components, or repeat services which prove to be defective in workmanship or material within product specific warranty periods or expiration dates and which our examination shall disclose to our satisfaction to be defective in such. This warranty is expressly in lieu of all other warranties, expressed or implied, as to description, quality, merchantability, fitness for any particular purpose, productiveness, or any other remedies, warranties, guarantees or liabilities, expressed or implied, arising by law or otherwise, and it shall have no liability for any lost profits or damage, direct, indirect or otherwise, to person or property, in connection with the use of any of its products or services. This warranty shall not be extended or varied except by written instrument signed by an authorized representative of NeoVentures Biotechnology Inc.

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