QUALICHEK™ Aflatoxin B1 Immunoaffinity Column

REF : KBIC1002-50

Designed and Developed as per AOAC Official Methods of Analysis Guidelines (Method AOAC 991.31 and others) and European Standards (EN 15851:2010).

Ver 1.1

RUO

For Use in Food and Feed Testing. Not For Use In Human or Animal Consumption.

Immunoaffinity Column for the use in Quantitative Determination of Aflatoxin B1 in cereals and feed.

RUO	**For Research Use Only**	**REF**	**Catalog Number**
	Store At	**LOT**	**Batch Code**
	Manufactured By		**Biological Risk**
	Expiry Date		**Consult Operating Instructions**

For Research Use Only.Purchase does not include or carry the right to resell or transfer this product either as a stand-alone product or as a component of another product. Any use of this product other than the permitted use without the express written authorization of KRISHGEN BioSystems is strictly prohibited.

REF KBIC1002-50 Σ 50 columns

KRISHGEN BioSystems
For US / Europe: toll free +1(888)-970-0827 tel: +1(562)-568-500
For Asia / India: tel: +91(22)-49198700
Email: sales@krishgen.com

Introduction:

Aflatoxin B1 (AFB1) is determined in food and feed using Qualichek™ Immunoaffinity Column (Immunoaffinity Chromotography (Qualichek™ IAC)) based on monoclonal antibodies bonded to agarose gel in combination with quantification with HPLC (High Performance Liquid Chromotography).

This method of content determination of aflatoxin B1 combines the high selectivity of an immunoaffinity column (IAC) with its potential to concentrate eluate with the additional step of purification by HPLC column.

Intended Use:

The QUALICHEK™ Aflatoxin B1 Immunoaffinity Column is used for determination of Aflatoxin B1 in cereals and feed with downstream testing by HPLC. The product is used for Aflatoxin B1 purification in cereal and feed samples prior to HPLC, HPLC-MS\MS or Specialized Rapid Tester. It reduces matrix interference and improves analysis accuracy.

Principle:

The columns are based on antigen-antibody binding reaction. Aflatoxin B1 monoclonal antibody is coupled to agarose gel material. After extracting, filtering and diluting Aflatoxin B1 in sample, sample extraction solution is slowly passed through the QUALICHEK™ IAC. Aflatoxin B1 in sample extraction solution combines with specific monoclonal antibody; leaving impurities which are washed away. Finally, Aflatoxin B1 compound is eluted to get purified Aflatoxin B1 by using methanol.

Materials Provided:

1. QUALICHEK™ Immunoaffinity Column – 50 columns
2. Instruction Manual

Materials, Not Provided:

1. ph Meter
2. Pipettes
3. Collection tray, tubes
4. Timer, watch
5. Acetonitrile
6. Purified Water
7. NaCl
8. K2HPO4
9. NaH2PO4
10. Tween 20

Reagent Preparation:

1. 90% acetonitrile (V:V) - Take 900 ml acetonitrile, then add 100 ml purified water.
2. PBST: Take 8 gms NaCl, 0.2 gm KCl, 0.2 gm K2HPO4, 1.2 gms NaH2PO4, then mix with 800 ml purified water. Later, add 1 ml Tween 20 and mix well. After that, adjust pH value to 7.4. Post settling, readjust final volume to 100 ml.

Sample preparation and purification:

A. General Solid Sample (Cereals and their products, Nuts and Seeds), Vegetable Oil

1. Take 5 gms pulverized sample. Add 25 ml 90% acetonitrile. Mix them in a homogenizer for 2.0 mins or vibrate for 30.0 mins and extract the sample.
2. Centrifuge at 5000 rpm for 5.0 minutes or filter through rapid qualitative filter paper. Later, take 5 ml filtrate, and add 25 ml water or PBST to dilute. Mix well.

3. For turbid solution, filter by glass fibre filter paper (or centrifuge it at 5000 rpm for 5.0 mins). Collect the filtrate.
4. Take 10 ml of the filtrate.

B. Sauce, Vinegar

1. Take 5 gms sample, then make it at constant volume 25 ml with acetonitrile. Mix them in a homogenizer for 2.0 mins or vibrate for 30 mins and extract the sample.
2. Centrifuge at 5000 rpm for 5.0 mins or filter through rapid qualitative filter paper. Later, take 5 ml filtrate, and add 25 ml water or PBST to dilute and mix well.
3. For turbid solution, filter by glass fiber filter paper (or centrifuge it at 5000 rpm for 5 mins), then collect filtrate.
4. Take 10 ml filtrate.

C. Infant Formula Food and Supplementary Food

1. Take 1 gm sample (accuracy 0.001 gm) into 50 ml Centrifuge. Tube, then add 4 ml hot water at 50°C. Use Vortex Mixer to mix well. If the milk powder cannot be completely dissolved, place the centrifuge tube in a 50°C water bath, and take the milk powder after it is completely dissolved.
2. Wait until the solution cooled down to 20°C, then add 10 ml methanol. Use Vortex Mixer for 3.0 mins.
3. Centrifuge at 4000 rpm for ten mins or filter through rapid qualitative filter paper.
4. Later, take 7 ml supernatant or treated solution, and then add 40 ml, water or PBST to dilute. Mix well.

Note:

1. For sample extraction solution (pH<6 or pH>8), it is necessary to adjust the pH value to neutral and use a glass fiber filter paper/quantitative filter paper.
2. For samples that are difficult to filter because of turbidity, centrifuge such samples for separation.

QUALICHEK™ Immunoaffinity Column Purification:

Note: Do not let column liquid drain dry before step 4.

1. Remove Qualichek™ IAC from the carton, and pierce its upward plug with 10 ml injector.
2. Fix Qualichek™ IAC on Pump Flow Operation Rack or Solid Phase Extraction Apparatus.
3. Add appropriate amount of treated sample filtrate into injector, and then remove Qualichek™ IAC downward plug.
4. Adjust flow rate to 1-1.5 ml / min.
5. After draining the liquid, wash it with 15 ml water or PBST at flow rate of 1-1.5 ml / min until 2~3 ml air passes through the column to ensure that there is no residual liquid in the column,
6. Elute with 1 ml methanol at flow rate of 1 ml/min, and use sample bottle / glass tube to collect the eluent.
7. Filter by 0.22 um organic phase filter head, to obtain purified Aflatoxin.

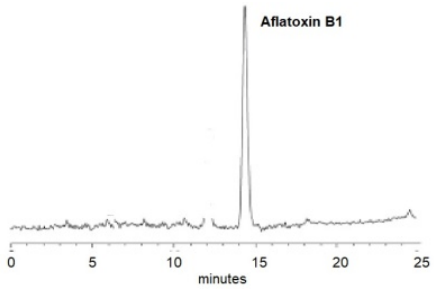
Sample Dilution Times:

General Solid Sample (Cereals and their products, Nuts and Seeds), Vegetable Oil, Sauce, Vinegar: 3
Infant Formula Food and Supplementary Food: 2

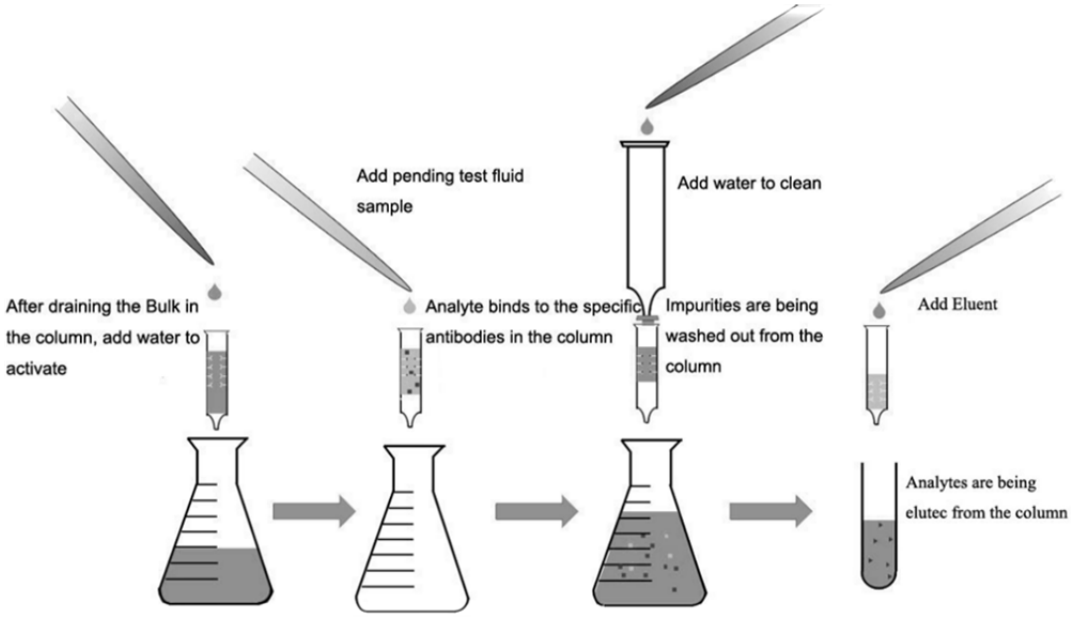
HPLC:

Column: C18, 5 um, 4.6 mm × 250 mm
Mobile Phase A: Methanol: Water is 45:55 (v:v)
Flow Rate: 1.0 ml/min
Detection Wavelength: Excitation Wavelength 360 nm, Emission Wavelength 440 nm
Sample Loading Quantity: 10 ul
Temperature: 40°C

Typical Chromatogram - Afaltoxin B1



Immunoaffinity Column Operation Diagram



Performance Characteristics:

Recoveries:
85%~110%

Column Capacity:
≥200 ng/vial

Column Gel:
Agarose Gel

Note:

1. Aflatoxin B1 is harmful to human, so please wear gloves while operation. All glassware exposed to standard/sample should be soaked overnight with 5% sodium hypochlorite solution.
2. Do not use expired Qualichek™ IAC.
3. Qualichek™ IAC should be stored at 2~8℃. Do not freeze.
4. Equilibrate Qualichek™ IAC at room temperature (25℃) for half an hour before use.
5. If Aflatoxin B1 content in sample is higher than column capacity, please decrease sample loading volume accordingly.
6. Adjust sample amount and extracting solution volume in proportion according to actual situation. It is recommended to take 5 gms sample in minimum.
7. Do not leave the Qualichek™ IAC in a dry state for a long time during the purification process.

LIMITED WARRANTY

Krishgen Biosystems does not warrant against damages or defects arising in shipping or handling, or out of accident or improper or abnormal use of the Products; against defects in products or components not manufactured by Krishgen Biosystems, or against damages resulting from such non-Krishgen Biosystems made products or components. Krishgen Biosystems passes on to customer the warranty it received (if any) from the maker thereof of such non Krishgen made products or components. This warranty also does not apply to Products to which changes or modifications have been made or attempted by persons other than pursuant to written authorization by Krishgen Biosystems.

THIS WARRANTY IS EXCLUSIVE. The sole and exclusive obligation of Krishgen Biosystems shall be to repair or replace the defective Products in the manner and for the period provided above. Krishgen Biosystems shall not have any other obligation with respect to the Products or any part thereof, whether based on contract, tort, and strict liability or otherwise. Under no circumstances, whether based on this Limited Warranty or otherwise, shall Krishgen Biosystems be liable for incidental, special, or consequential damages.

This Limited Warranty states the entire obligation of Krishgen Biosystems with respect to the Products. If any part of this Limited Warranty is determined to be void or illegal, the remainder shall remain in full force and effect.

Krishgen Biosystems. 2022

THANK YOU FOR USING A KRISHGEN PRODUCT!