Intended Use
The QuickTox Kit for QuickScan Aflatoxin FREE is designed to quickly provide quantitative results for the presence of total aflatoxins. Refer to the below table for specific detection ranges, which are dependent upon matrix group and dilution.

<table>
<thead>
<tr>
<th>Matrix Group (MG)</th>
<th>LOD</th>
<th>Maximum Reported Value of Base Range</th>
<th>Range with Dilution</th>
</tr>
</thead>
<tbody>
<tr>
<td>MG1-MG8</td>
<td>2.5-2.7 ppb¹</td>
<td>30 ppb</td>
<td>&gt;30-100 ppb</td>
</tr>
<tr>
<td>MG9 – Corn (high sensitivity)⁴</td>
<td>1.5 ppb</td>
<td>20 ppb</td>
<td>&gt;20-100 ppb</td>
</tr>
<tr>
<td>MG10-MG12</td>
<td>7.5 ppb</td>
<td>99 ppb</td>
<td>&gt;99-300 ppb¹</td>
</tr>
<tr>
<td>MG13-MG16</td>
<td>2.7 ppb</td>
<td>30 ppb</td>
<td>&gt;30-100 ppb</td>
</tr>
<tr>
<td>MG17 – Peanut Seed (high sensitivity)</td>
<td>2.5 ppb</td>
<td>30 ppb</td>
<td>N/A</td>
</tr>
</tbody>
</table>

¹ Matrix Group Dependent
² Dilution is performed only for samples with results above the base range. After running a diluted sample, selecting 1:A from the Dilution tab in the QuickScan results window adjusts for the dilution factor.
³ MG3 reports results down to ‘0’, with an LOD of 2.7 ppb. Do not assume accuracy for results reported below the LOD.
⁴ MG9 – Corn (high sensitivity) reports results down to ‘0’, with an LOD of 1.5 ppb. Do not assume accuracy for results reported below the LOD.

This assay has been certified up to 100 ppb as a Performance Tested MethodSM, #071502 by the AOAC Research Institute for use in corn and wheat using aqueous extraction (EB17), as well as in barley, oats, sorghum, whole peanut, peanut hull and peanut seed using organic solvent extraction. This assay has also been certified up to 30 ppb for the Peanut Seed (high sensitivity) protocol using the organic solvent extraction.

How the Test Works
A composite sample is first collected, ground, and extracted to solubilize any aflatoxin present. The extract is further diluted into Buffer before being run on the QuickTox test strip.

Each QuickTox Strip has an absorbent pad at each end. The sample extract travels up the test strip and is absorbed into the larger pad at the top of the strip. At the end of the reaction time, the strip is cut at the top of the arrow tape, the bottom pads are discarded, and the strip is inserted into the QuickScan reader to obtain quantitative results.

Matrix specific extractions and analysis protocols are chosen for accuracy and precision. Each matrix is assigned to a Matrix Group (MG). Each MG has a common standard curve, Limit of Detection (LOD), and maximum reported value. When the user selects the MG during testing, the QuickScan System software reads the test strip, retrieves information encoded in the strip’s barcode and on the Multi-Matrix Barcode Card (MMBC), and uses the appropriate curve to obtain a result for the matrix being tested.

Matrices

Note: Scanning the Multi-Matrix Barcode Card once per kit lot is required. The QuickScan software will prompt users to select a Matrix Group (MG) before proceeding to the result screen. If you only plan to test matrices within the MG1 group (Corn, Brown Rice and Wheat), scan the side of the MMBC card that only has the MG1 barcode. This allows the software to skip the step which prompts users to select a Matrix Group.
QuickTox Kit for QuickScan Aflatoxin FREE
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- Corn
- Brown Rice
- Wheat

EB17 Buffer Extraction

**SET A**
PROCEDURES: PAGE 5

- Barley
- Coconut Meal
- Corn (high sensitivity)
- Corn Flour
- Corn Germ
- Corn Gluten Meal
- Cottonseed (delinted)

**SET B**
PROCEDURES: PAGE 6

- Rice Bran
- Rice Hulls
- Rye, Whole
- Sorghum
- Soybean Meal

- Corn Germ Meal
- Corn Gluten Feed
- Corn Silage
- Cottonseed Meal

**SET C**
PROCEDURES: PAGE 7

50% Ethanol Extraction

- DDGS
- Hominy Feed
- Oats
- Rice, Black Glutinous
- Rice, Rough
- Rice, White

- Barley
- Coconut Meal
- Corn (high sensitivity)
- Corn Flour
- Corn Germ
- Corn Gluten Meal
- Cottonseed (delinted)

**SET D**
PROCEDURES: PAGE 8

- DDGS
- Hominy Feed
- Oats
- Rice, Black Glutinous
- Rice, Rough
- Rice, White

- Barley
- Coconut Meal
- Corn (high sensitivity)
- Corn Flour
- Corn Germ
- Corn Gluten Meal
- Cottonseed (delinted)

**SET E**
PROCEDURES: PAGE 9

50% Ethanol Extraction

- Rice Bran
- Rice Hulls
- Rye, Whole
- Sorghum
- Soybean Meal

- Corn Germ Meal
- Corn Gluten Feed
- Corn Silage
- Cottonseed Meal

- Barley
- Coconut Meal
- Corn (high sensitivity)
- Corn Flour
- Corn Germ
- Corn Gluten Meal
- Cottonseed (delinted)

**SET F**
PROCEDURES: PAGE 10

50% Ethanol Extraction

- Rice Bran
- Rice Hulls
- Rye, Whole
- Sorghum
- Soybean Meal

**Items Not Provided:**
- QuickScan System*
- Bunn grinder or equivalent
- Coffee grinder or equivalent
- 20 mesh screen
- Extraction cups with lids (for 25g samples)* or other suitable vessels for sample extraction*
- Graduated cylinder*
- Orbital/rotary shaker
- Pipette to deliver 100 µL*
- Approved coffee filters*
- Tubes and pipettes for centrifugation*
- Microcentrifuge*
- Vials for additional dilution of high samples*
- Pipette to deliver larger volumes for dilutions
- Timer
- Scissors
- Distilled, deionized or bottled water
- Extraction bags (small or large depending on sample size, for cottonseed meal and corn gluten feed)
- Ethanol 50%* (Reagent Alcohol, for some matrices)
- Ethanol 80%* (Reagent Alcohol, for some matrices)
- Acetonitrile, 50%* and/or 84%* (for some matrices)
- Acetic acid, 7% (for hazelnuts)
- DB5 Buffer (additional, for some matrices)*
- Table salt, non-iodized (for peanut matrices)
- 7 mesh screen (for peanut seed and whole peanut)
- 12 X 75mm polypropylene tubes* (High Sensitivity Peanut protocol only)
- Incubator* (High Sensitivity Peanut protocol only)

*Available as Accessories →

**Available Accessories:**

<table>
<thead>
<tr>
<th>Item</th>
<th>Cat. No.</th>
<th>Part #</th>
</tr>
</thead>
<tbody>
<tr>
<td>QuickScan™ System</td>
<td>ACC 131</td>
<td>100504 + 11222</td>
</tr>
<tr>
<td>50 Sample cups/lids (for 25g samples) Please note: if using these cups with an acetonitrile extraction, they may leak; seal covers onto cups with Parafilm or similar sealant</td>
<td>ACC 012-50</td>
<td>11224</td>
</tr>
<tr>
<td>Graduated cylinder (100 mL)</td>
<td>ACC 068</td>
<td>11207</td>
</tr>
<tr>
<td>MiniPet pipette 100 µL (one/location free)</td>
<td>ACC 041</td>
<td>11202</td>
</tr>
<tr>
<td>Coffee filters (100)</td>
<td>ACC 083</td>
<td>11434</td>
</tr>
<tr>
<td>Centrifugation Set: Disposables for 50 tests</td>
<td>ACC 010</td>
<td>11214</td>
</tr>
<tr>
<td>Microcentrifuge</td>
<td>ACC 064 E</td>
<td>11204</td>
</tr>
<tr>
<td>50g Sample Extraction Set</td>
<td>ACC 035</td>
<td>11216</td>
</tr>
<tr>
<td>Additional Powder Packets and Sample Extraction bags</td>
<td></td>
<td></td>
</tr>
<tr>
<td>FREE Dilution Set: Disposables and Extraction Powder Packets for 100 Dilutions</td>
<td>ACC 034</td>
<td>11215</td>
</tr>
<tr>
<td>QuickTox Dilution Set: Tips + vials for 100 dilutions for testing samples above base range</td>
<td>ACC 080</td>
<td>11219</td>
</tr>
<tr>
<td>50% Ethanol</td>
<td>ACC E26902-1X</td>
<td>11156</td>
</tr>
<tr>
<td>80% Ethanol</td>
<td>ACC EE23805</td>
<td>11963</td>
</tr>
<tr>
<td>84% Acetonitrile</td>
<td>ACC-EE19702-4L</td>
<td>11932</td>
</tr>
<tr>
<td>50% Acetonitrile</td>
<td>ACC-LC104604-4L</td>
<td>11962</td>
</tr>
<tr>
<td>DB5 Buffer</td>
<td>KR-266-7</td>
<td>11665</td>
</tr>
<tr>
<td>Additional Buffer needed for matrices requiring &gt; 100 µL per Strip</td>
<td></td>
<td></td>
</tr>
<tr>
<td>12 X 75mm poly tubes</td>
<td>20-0128</td>
<td>12198</td>
</tr>
<tr>
<td>Incubator</td>
<td>ACC-BSH301</td>
<td>12458</td>
</tr>
</tbody>
</table>

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Precautions – Read First!

SAFETY

1. Disposal of aflatoxin-contaminated materials.
   a. Follow your facility’s safety procedures for disposal of samples and extracts potentially containing or known to contain aflatoxin.

2. EB17 Extraction Powder is flammable and an irritant. See attached Safety Data Sheet.
   a. Avoid inhaling powder or contact with the skin, eyes, or clothing. Wear personal protective equipment including safety glasses, gloves, mask and lab coat when handling. Keep powder away from heat, sparks and open flame.
   b. Observe any applicable regulations when disposing of extracted samples and kit reagents.
   c. Do not treat either the EB17 extracts or the EB17 extraction labware with bleach; the Extraction Packet powder is incompatible with strong oxidizers.

3. Ethanol and acetonitrile are flammable and toxic.
   a. Avoid inhaling vapors or contact with the skin, eyes, or clothing. Wear personal protective equipment including safety glasses, nitrile gloves (not latex), a vapor mask and a lab coat when handling. Keep containers tightly closed and away from heat, sparks and open flame.
   b. Observe any applicable regulations when disposing of samples and kit reagents.

4. Acetonitrile may leak.
   a. Use caution when sealing extraction cups, assure a tight seal.
   b. To avoid leaks when using Sample Cups (ACC-012), wrap Parafilm® or similar product around the outside cup threads in the direction of the threads before screwing on cap.

GENERAL

1. The intended user should read the entire product instructions, including all safety precautions, before use of this kit. The operator should be capable of using common testing equipment including an appropriate grinder or mill, pipettes, graduated cylinders, etc. Training on use of this product and the QuickScan System is available from EnviroLogix.

2. Test strip canisters are desiccated; before opening canisters, ensure they have warmed to room temperature. After removing test strips, reseal the canister immediately. Avoid bending test strips.

3. Ensure all samples, extraction reagents (including water), test strips, and Buffer are at room temperature before use.

4. Test extracts within 5 minutes of diluting with Buffer for optimal performance.

Sample Preparation

1. Collect a composite sample according to your own sampling plan or USDA/GIPSA guidelines. Consult USDA/GIPSA reference documents to help design a plan that fits your needs. Contact Technical Support for more information. Note, Corn Silage procedure was qualified using samples with a moisture content in the range of 50-70%, which is a typical range for this matrix.

2. Unless noted, grind samples to provide a consistency such that 95% passes through a 20 mesh sieve.
   Note, Wheat: Grinding wheat too finely may impact flow and accuracy. Contact Technical Support for information.
   Note, Peanut: The speed of the grinder needs to be controlled to prevent sample overheating and oil release with peanut seed and whole peanut. An optimal finished grind allows about 90% to pass through a 7 mesh sieve.
   Note, Corn Silage: Must use a coffee grinder or equivalent, for 1 minute, to achieve the correct grind consistency.

3. Mix ground material thoroughly before sub-sampling, to minimize variability.

4. Weigh 25g or 50g samples into containers that will allow enough head room for the liquid to move forcefully when shaken vigorously.

Sample Clarification

Depending on the sample matrix, there may be multiple acceptable methods for removing particulate from the extract.

<table>
<thead>
<tr>
<th>Centrifugation</th>
<th>Filtration</th>
<th>Settling</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Fill a microcentrifuge tube with extract.</td>
<td>1. Add an approved coffee filter (e.g. BUNN Part #BUNBCF100B) to a clean vessel.</td>
<td>1. Allow the sample to sit undisturbed until a top layer forms that can easily be pipetted.</td>
</tr>
<tr>
<td>2. Centrifuge for the specified time at 2000 x g (ref, not rpm).</td>
<td>2. Pour extract into the filter.</td>
<td>2. Use the top layer of extract.</td>
</tr>
<tr>
<td>3. Use the top layer of extract.</td>
<td>3. Pull back the filter to access the filtered extract.</td>
<td></td>
</tr>
</tbody>
</table>

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Range with Dilution

For testing samples at levels greater than the assay's base range

1. If after running and reading the test, the initial result is greater than the upper end of the Base Range, samples can be diluted and retested to extend quantitation (see table on p.1).

2. Combine extract with the appropriate extraction reagent (EB17 Dilution Solution, Ethanol, Acetonitrile) to create a 1:6 dilution. Example: 1 part clarified extract + 5 parts diluent; 100 µL + 500 µL. Measure carefully and mix well.
   Note: for EB17-extracted matrices, a liquid EB17 Dilution Solution must be prepared. Mix 1 packet EB17 powder with 300 mL of water and mix well; Dilution Solution mixture will appear cloudy. It may be stored after mixing for up to 30 days at room temperature. Re-suspend solution before each use.

3. Rerun assay as before, adding Buffer + diluted extract into the reaction vial, then adding a new strip for the time specified. Example: for corn, pipette 100 µL DB5 + 100 µL of the extract diluted with Dilution Solution into a new vial, add a new test strip, and wait 4 minutes for test results.

4. In the QuickScan Results Screen, select 1:A under the dilution tab (dropdown menu). The System will calculate and record the aflatoxin level in diluted samples.
   Note: Dilution accessory sets are available, see items ACC-080 and ACC-034 (includes EB17 packets).

Use of the QuickScan System

Detailed instructions for use of the QuickScan System are supplied with each unit, and can also be found at www.envirologix.com/quickscan. The lot-specific Multi-Matrix Barcode Card must be scanned into the system prior to testing.

In summary, a strip is inserted face down in the carrier with the barcoded end closest to the handle. The carrier is inserted into the reader and the strips are read by touching or clicking on the “Read Test” area of the screen. The “Select Matrix Groups” screen will appear. Select the group that displays the matrix run. Results are then recorded in an electronic worksheet, allowing each user to report and track data easily.

Kit Storage

This QuickTox Kit should be stored refrigerated. Note the shelf life on the kit box. Prolonged exposure to high temperatures may adversely affect the test results; protect all components from extreme hot or cold temperatures. Do not leave in direct sunlight or in a vehicle. Do not open the desiccated canister until ready to use the strips.

Cross-reactivity

The following mycotoxins have been tested with this kit and no false positive results occurred at the 200 ppm level: DON (deoxynivalenol), Fumonisin B1, Ochratoxin A, Zearalenone.

Precautions and Notes

- This product is currently not applicable for use in testing any other crops beyond those specified in this Product Insert.
- This assay is calibrated against reference samples supplied by Trilogy Analytical Laboratory, Washington, MO, and other vendors and associated HPLC data. Performance in other sample matrices has been validated using fortified samples.
- As with all screening tests, it is recommended that results be confirmed by an alternate method when necessary.
- The assay has been optimized for use with the protocols provided in the kit. Deviation from these protocols may invalidate the results of the test. Proper mixing, timing, and accurate pipetting are essential to accurate results.
- The results generated through the proper use of this diagnostic tool reflect the condition of the working sample directly tested. Extrapolation as to the condition of the originating lot, from which the working sample was derived, should be based on sound sampling procedures and statistical calculations which address random sampling effects, non-random sampling effects and assay system uncertainty. A negative result obtained when properly testing the working sample does not necessarily mean the originating lot is entirely negative for the analyte in question.
SET A Procedures: EB17 Aqueous Matrices

Matrices: • Brown Rice • Corn (EB17 Extraction) • Wheat

- Review Sample Preparation on page 3 for grinding consistency and notes.
- Use distilled, deionized, or flat (non-carbonated) bottled water. Drinkable (potable) tap water may be used, with customer validation of water supply. Contact Technical Support to purchase a control set and protocol that can be used to verify your water supply.
- If testing 50-gram samples, additional EB17 Buffer packets are required (order Catalog No. ACC-035)

Sample Extraction

<table>
<thead>
<tr>
<th>25g Samples</th>
<th>50g Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Corn, Brown Rice</strong></td>
<td></td>
</tr>
<tr>
<td>1. Add 1 packet of EB17 to sample</td>
<td>1. Add 2 packets of EB17 to sample</td>
</tr>
<tr>
<td>2. Add 75 mL water</td>
<td>2. Dry Blend EB17 into sample</td>
</tr>
<tr>
<td></td>
<td>3. Add 150 mL water</td>
</tr>
<tr>
<td><strong>Wheat</strong></td>
<td></td>
</tr>
<tr>
<td>1. Add 1 packet EB17 to sample</td>
<td>1. Add 150 mL water, wet thoroughly</td>
</tr>
<tr>
<td>2. Add 75 mL water</td>
<td>2. Add 2 packets of EB17</td>
</tr>
</tbody>
</table>

**Shake:** choose mechanical shaker or hand shaking

**Clarify Extract:** choose centrifuge or filter

- Do not filter wheat samples, centrifuge only.

**Centrifuge:** 30 seconds at 2000 x g (ref, not rpm)

**Filter:** Pour through approved coffee filter (ACC-083)

Combine Buffer and Extract, then Run Test Strips

1. Add 100 µL DB5 to the reaction vial (discard tip)
2. Add 100 µL clarified extract to the reaction vial
3. Mix thoroughly with extract pipette tip, discard tip
4. Add test strip to vial, arrows down, wait for run time
   4 minutes: Corn and Brown Rice
   5 minutes: Wheat
5. Immediately cut strips at the top of the arrow tape (discard bottom pads)
6. Insert strip, barcode face down, into QuickScan Reader
7. If prompted, select “MG1 – Brown Rice, Corn, Wheat”

**TIPS!**

**Get Complete Extraction**
- Fully wet samples before shaking
- Assure liquid is moving forcefully though the sample while shaking

**For Best Performance**
- Pipette up and down while mixing
- Do not reuse diluted samples
- Read strips promptly after run time

Avoid Contamination
- Use a new reaction vial per test
- Keep DB5 capped, when possible
- Use new pipette tips for each step

**TABLE A: EB17-Extracted Matrix Summary Guide**

<table>
<thead>
<tr>
<th>Matrix</th>
<th>LOD (ppb)</th>
<th>First</th>
<th>Second</th>
<th>Third</th>
<th>Shake</th>
<th>Clarify</th>
<th>Reaction Vial</th>
<th>Run</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brown Rice</td>
<td>2.7</td>
<td>25g</td>
<td>1 x EB17</td>
<td>75 mL water</td>
<td>1 min – shaker</td>
<td>Filter or</td>
<td>100 µL DB5</td>
<td>4 min</td>
</tr>
<tr>
<td></td>
<td></td>
<td>50g</td>
<td>2 x EB17, dry blend</td>
<td>150 mL water</td>
<td>or by hand</td>
<td>Centrifuge</td>
<td>100 µL DB5 extract</td>
<td></td>
</tr>
<tr>
<td>Corn</td>
<td></td>
<td>25g</td>
<td>1 x EB17</td>
<td>75 mL water</td>
<td>1 min – shaker</td>
<td>Filter or</td>
<td>100 µL DB5</td>
<td>4 min</td>
</tr>
<tr>
<td></td>
<td></td>
<td>50g</td>
<td>2 x EB17, dry blend</td>
<td>150 mL water</td>
<td>or by hand</td>
<td>Centrifuge</td>
<td>100 µL DB5 extract</td>
<td></td>
</tr>
<tr>
<td>Wheat</td>
<td></td>
<td>25g</td>
<td>1 x EB17</td>
<td>75 mL water</td>
<td>1 min – shaker</td>
<td>Centrifuge</td>
<td>100 µL DB5</td>
<td>5 min</td>
</tr>
<tr>
<td></td>
<td></td>
<td>50g</td>
<td>150 mL water</td>
<td>2 x EB17</td>
<td>or by hand</td>
<td>Centrifuge</td>
<td>100 µL DB5 extract</td>
<td></td>
</tr>
</tbody>
</table>

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## SET B Procedures: Additional Matrices

<table>
<thead>
<tr>
<th>Matrices:</th>
<th>Corn Flour</th>
<th>DDGS</th>
<th>Rice, Rough</th>
<th>Rice Hulls</th>
</tr>
</thead>
<tbody>
<tr>
<td>Barley</td>
<td>Corn Germ</td>
<td>Hominy Feed</td>
<td>Rice, White</td>
<td>Rye, Whole</td>
</tr>
<tr>
<td>Coconut Meal</td>
<td>Corn Gluten Meal</td>
<td>Oats</td>
<td>Rice, White Glutinous</td>
<td>Sorghum</td>
</tr>
<tr>
<td>Corn (high sens)</td>
<td>Cottonseed (delinted)</td>
<td>Rice, Black Glutinous</td>
<td>Rice Bran</td>
<td>Soybean Meal</td>
</tr>
</tbody>
</table>

Review Sample Preparation on page 3 for grinding consistency and notes

### Sample Extraction:
Consult TABLE B below to determine if 2x or 4x 50% ethanol extraction is required

<table>
<thead>
<tr>
<th>25g Samples</th>
<th>50g Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>2x ethanol</strong></td>
<td>Add 50 mL 50% ethanol to sample</td>
</tr>
<tr>
<td><strong>4x ethanol</strong></td>
<td>Add 100 mL 50% ethanol to sample</td>
</tr>
</tbody>
</table>

**Shake:** choose mechanical shaker or hand shaking

**Shaker Table:** mix at highest speed for 1 minute

**By Hand:** shake vigorously for 2 minutes

*For oats, centrifuged immediately after shaking, or paste will form

**Clarify Extract:** Centrifuged for 1 minute at 2000 x g (rcf, not rpm)

**Combine Buffer and Extract, then Run Test Strips**

1. Consult TABLE B to determine DB5 and extract volume
2. Add DB5 to the reaction vial (discard tip)
3. Add clarified extract to the reaction vial
4. Mix thoroughly with extract pipette tip, discard tip
5. Add test strip to vial, arrows down
6. Wait 5 minutes (run time). For cottonseed, wait 7 minutes.
7. Immediately cut strips at the top of the arrow tape (discard bottom pads)
8. Insert strip, barcode face down, into QuickScan Reader
9. When prompted, select Matrix Group for the matrix being tested

### TABLE B: 50% Ethanol-Extracted Matrix Summary Guide

<table>
<thead>
<tr>
<th>Matrix Group</th>
<th>Matrix Group</th>
<th>LOD (ppb)</th>
<th>Ethanol Ratio</th>
<th>Shake</th>
<th>Clarify</th>
<th>DB5 Volume</th>
<th>Extract Volume</th>
<th>Run Time</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Corn High Sensitivity</em></td>
<td>MG9</td>
<td>1.5</td>
<td>2x</td>
<td>300 µL</td>
<td>200 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cottonseed, Delinted</td>
<td>MG2</td>
<td>2.5</td>
<td>4x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>7 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Barley</td>
<td>MG8</td>
<td>2.7</td>
<td>2x</td>
<td>200 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Corn Flour</td>
<td>MG8</td>
<td>2.5</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Oats</td>
<td>MG7</td>
<td>2.7</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice, Rough</td>
<td>MG7</td>
<td>2.5</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rye, Whole</td>
<td>MG6</td>
<td>2.7</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sorghum</td>
<td>MG7</td>
<td>2.7</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Soybean Meal</td>
<td>MG8</td>
<td>2.5</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Coconut Meal</td>
<td>MG3*</td>
<td>2.7</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Corn Germ</td>
<td>MG2</td>
<td>2.5</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Corn Gluten Meal</td>
<td>MG3*</td>
<td>2.7</td>
<td>4x</td>
<td>200 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>DDGS</td>
<td>MG2</td>
<td>2.5</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hominy Feed</td>
<td>MG3*</td>
<td>2.7</td>
<td>4x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice, Black Glutinous</td>
<td>MG3*</td>
<td>2.7</td>
<td>4x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice Bran</td>
<td>MG2</td>
<td>2.5</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice Hulls</td>
<td>MG16</td>
<td>2.7</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice, White</td>
<td>MG15</td>
<td>2.7</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice, White Glutinous</td>
<td>MG14</td>
<td>2.7</td>
<td>2x</td>
<td>100 µL</td>
<td>100 µL</td>
<td>5 min</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Results reported down to "0"; however, do not assume accuracy for results reported below the assay's LOD

---

**TIPS!**

**Get Complete Extraction**
- Fully wet samples before shaking
- Assure liquid is moving forcefully though the sample while shaking

**For Best Performance**
- Pipette up and down while mixing
- Do not reuse diluted samples
- Read strips promptly after run time

**Avoid Contamination**
- Use a new reaction vial per test
- Keep DB5 capped when possible
- Use new pipette tips for each step
SET C Procedures: Additional Matrices

**Matrices:**  • Corn Germ Meal  • Corn Gluten Feed  • Corn Silage  • Cottonseed Meal

Review Sample Preparation on page 3 for grinding consistency and notes

**Sample Extraction:** Add the appropriate solvent to the sample

<table>
<thead>
<tr>
<th></th>
<th>25g Samples</th>
<th>50g Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Corn Germ Meal</strong>, <strong>Corn Silage</strong></td>
<td>Add 50 mL 80% Ethanol</td>
<td>Add 100 mL 80% Ethanol</td>
</tr>
<tr>
<td><strong>Corn Gluten Feed</strong></td>
<td>Add 40 mL 84% Acetonitrile</td>
<td>Add 80 mL 84% Acetonitrile</td>
</tr>
<tr>
<td><strong>Cottonseed Meal</strong></td>
<td>Add 50 mL 50% Acetonitrile</td>
<td>Add 100 mL 50% Acetonitrile</td>
</tr>
</tbody>
</table>

**Shake:** choose mechanical shaker or hand shaking

| **Shaker Table:** mix at highest speed for 1 minute (Corn gluten feed, 2 minutes) | **By Hand:** shake vigorously for 2 minutes |

**Clarify Extract:** Let extract settle

- Corn Germ Meal: 2 minutes
- Corn Gluten Feed: 1 minute
- Cottonseed Meal: at least 2 minutes
- Corn Silage: Centrifuge 1 min at 2000 x g

**Combine Buffer and Extract, then Run Test Strips**

1. **Consult TABLE C** to determine DB5 and extract volume
   **Note, Corn Gluten Feed:** Pre-mix DB5 and extract in a clean vial. Add 200 µL pre-mix to reaction vial.
2. Add DB5 to the reaction vial (discard tip)
3. Add clarified extract to the reaction vial
4. Mix thoroughly with extract pipette tip, discard tip
5. Add test strip to vial, arrows down
6. Wait 5 minutes (run time)
7. Immediately cut strips at the top of the arrow tape (discard bottom pads)
8. Insert strip, barcode face down, into QuickScan Reader
9. When prompted, select Matrix Group for the matrix being tested

**TIPS!**

**Get Complete Extraction**
- Fully wet samples before shaking
- Assure liquid is moving forcefully though the sample while shaking

**For Best Performance**
- Pipette up and down while mixing
- Do not reuse diluted samples
- Read strips promptly after run time

**Avoid Contamination**
- Use a new reaction vial per test
- Keep DB5 capped when possible
- Use new pipette tips for each step

**TABLE C: Other Solvents Matrix Summary Guide**

<table>
<thead>
<tr>
<th>Matrix</th>
<th>Matrix Group</th>
<th>LOD (ppb)</th>
<th>Extraction</th>
<th>Shake</th>
<th>Clarify (Settle)</th>
<th>DB5 Volume</th>
<th>Extract Volume</th>
<th>Run Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn Germ Meal</td>
<td>MG6</td>
<td>2.7</td>
<td>2x, 80% Ethanol</td>
<td>1 min – shaker or 2 min – hand</td>
<td>2 min</td>
<td>200 µL</td>
<td>100 µL</td>
<td></td>
</tr>
<tr>
<td>Corn Gluten Feed</td>
<td>MG5</td>
<td>2.5</td>
<td>1.6x, 84% Acetonitrile*</td>
<td>2 min – shaker or 2 min – hand</td>
<td>1 min</td>
<td>Pre-mix 500 µL DB5 + 100 µL extract (test 200 µL)</td>
<td>5 min</td>
<td></td>
</tr>
<tr>
<td>Corn Silage</td>
<td>MG3**</td>
<td>2.7</td>
<td>2x, 80% Ethanol</td>
<td>1 min – shaker or 2 min – hand</td>
<td>Centrifuge 1 min at 2000 x g</td>
<td>200 µL</td>
<td>100 µL</td>
<td></td>
</tr>
<tr>
<td>Cottonseed Meal</td>
<td>MG4</td>
<td>2.5</td>
<td>2x, 50% Acetonitrile*</td>
<td>1 min – shaker or 2 min – hand</td>
<td>≥ 2 min</td>
<td>200 µL</td>
<td>100 µL</td>
<td></td>
</tr>
</tbody>
</table>

*Acetonitrile may leak; refer to page 3 for preventative measures.

**Results reported down to "0"; however, do not assume accuracy for results reported below the assay's LOD
SET D Procedures: Additional Matrices

**Matrices:**
- Peanut Hull
- Peanut Seed
- Whole Peanut

Review Sample Preparation on page 3 for grinding consistency and notes

**Sample Extraction**

Prepare a bulk salt water solution: (a) Add 29.4g of table salt (non-iodized) per 100 mL of bottled water.
(b) Mix well, until salt is in solution.

<table>
<thead>
<tr>
<th>25g Samples</th>
<th>50g Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>Create Slurry*</td>
<td>Add 20 mL salt water to sample</td>
</tr>
<tr>
<td></td>
<td>2. Mix well, stir slowly</td>
</tr>
<tr>
<td>Add Solvent</td>
<td>3. Add 75 mL 80% Ethanol</td>
</tr>
<tr>
<td></td>
<td>4. Make sure entire sample is wetted</td>
</tr>
</tbody>
</table>

*Note: Peanut hull slurry will not have the same consistency as peanut seed and whole peanut, it will be more of a dry mixture due to the absorbency of the matrix*

The above extraction protocol provides instruction for 25g and 50g samples. To customize sample size, keep salt water slurry and Ethanol ratios the same.

| Salt water: | 0.8 mL/g sample |
| Ethanol: | 3 mL/g sample |

**Example: 200 g sample**
- 160 mL salt water
- 600 µL 80% Ethanol

**Shake:** choose mechanical shaker or hand shaking

- **Shaker Table:** mix at highest speed for 1 minute
- **By Hand:** shake vigorously for 2 minutes

**Clarify Extract:**

Pour through an approved coffee filter (e.g. ACC-083). Mix the clarified extract well before testing.

**Combine Buffer and Extract, then Run Test Strips**

1. Consult TABLE D to determine DB5 and extract volume
2. Add DB5 to the reaction vial (discard tip)
3. Add clarified extract to the reaction vial
4. Mix thoroughly with extract pipette tip (discard tip)
5. Add test strip to vial, arrows down
6. Wait 4 minutes (run time)
7. Immediately cut strips at the top of the arrow tape (discard bottom pads)
8. Insert strip, barcode face down, into QuickScan reader
9. When prompted, select Matrix Group for the matrix being tested

**TABLE D: Peanut Matrix Summary Guide**

<table>
<thead>
<tr>
<th>Matrix</th>
<th>Matrix Group</th>
<th>LOD (ppb)</th>
<th>Slurry</th>
<th>Extract-ant</th>
<th>Shake</th>
<th>Clarify</th>
<th>DB5 Volume</th>
<th>Extract Volume</th>
<th>Run Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peanut Hull</td>
<td>MG12</td>
<td>25g: add 20 mL salt water</td>
<td>3x, 80% Ethanol</td>
<td>1 min – shaker or 2 min – by hand</td>
<td>Filter; mix well</td>
<td>200 µL</td>
<td>100 µL</td>
<td>4 min</td>
<td></td>
</tr>
<tr>
<td>Peanut Seed</td>
<td>MG10</td>
<td>7.5</td>
<td>50g: add 40 mL salt water</td>
<td></td>
<td></td>
<td>400 µL</td>
<td>100 µL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole Peanut</td>
<td>MG11</td>
<td>400 µL</td>
<td>100 µL</td>
<td>4 min</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**TIPS!**

**Get Complete Extraction**
- Fully wet samples before shaking
- Assure liquid is moving forcefully though the sample while shaking

**For Best Performance**
- Pipette up and down while mixing
- Do not reuse diluted samples
- Read strips promptly after run time

**Avoid Contamination**
- Use a new reaction vial per test
- Keep DB5 capped, when possible
- Use new pipette tips each step
SET E Procedures: Additional Matrices

**Matrices:**
- Hazelnut

Review Sample Preparation on page 3 for grinding consistency and notes

**Sample Extraction**

Prepare a bulk salt water solution: (a) Add 29.4g of table salt (non-iodized) per 100 mL of bottled water.
(b) Mix well, until salt is in solution.

<table>
<thead>
<tr>
<th>25g Samples</th>
<th>50g Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Create Slurry</strong></td>
<td></td>
</tr>
<tr>
<td>1. Add 20 mL salt water to sample</td>
<td>1. Add 40 mL salt water to sample</td>
</tr>
<tr>
<td>2. Mix well, stir slowly</td>
<td>2. Mix well, stir slowly</td>
</tr>
<tr>
<td><strong>Add Solvent</strong></td>
<td></td>
</tr>
<tr>
<td>3. Add 72 mL 80% Ethanol</td>
<td>3. Add 144 mL 80% Ethanol</td>
</tr>
<tr>
<td>4. Add 3 mL of 7% Acetic Acid</td>
<td>4. Add 6 mL of 7% Acetic Acid</td>
</tr>
<tr>
<td>5. Make sure entire sample is wetted</td>
<td>5. Make sure entire sample is wetted</td>
</tr>
</tbody>
</table>

*Note: Commercial vinegar with 7% acetic acid may be used.*

The above extraction protocol provides instruction for 25g and 50g samples. To customize sample size, keep salt water slurry, Ethanol and Acetic Acid ratios the same.

<table>
<thead>
<tr>
<th>Salt water: 0.8 mL/g sample</th>
<th>Example: 200 g sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>80% Ethanol: 2.88 mL/g sample</td>
<td>- 160 mL salt water</td>
</tr>
<tr>
<td>7% Acetic Acid: 0.12 mL/g sample</td>
<td>- 576 mL 80% Ethanol</td>
</tr>
<tr>
<td>- 24 mL 7% Acetic Acid</td>
<td></td>
</tr>
</tbody>
</table>

**Shake:** choose mechanical shaker or hand shaking

**Shaker Table:** mix at highest speed for 1 minute

**By Hand:** shake vigorously for 2 minutes

**Clarify Extract:**

Pour through an approved coffee filter (e.g. ACC-083). Mix the clarified extract well before testing.

**Combine Buffer and Extract, then Run Test Strips**

1. Consult TABLE E to determine DB5 and extract volume
2. Add DB5 to the reaction vial (discard tip)
3. Add clarified extract to the reaction vial
4. Mix thoroughly with extract pipette tip (discard tip)
5. Add test strip to vial, arrows down
6. Wait 4 minutes (run time)
7. Immediately cut strips at the top of the arrow tape (discard bottom pads)
8. Insert strip, barcode face down, into QuickScan reader
9. When prompted, select Matrix Group for the matrix being tested

**TABLE E: Matrix Summary Guide**

<table>
<thead>
<tr>
<th>Matrix</th>
<th>Matrix Group</th>
<th>LOD (ppb)</th>
<th>Slurry</th>
<th>Extractant</th>
<th>Shake</th>
<th>Clarify</th>
<th>DB5 Volume</th>
<th>Extract Volume</th>
<th>Run Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hazelnut Seed</td>
<td>MG10</td>
<td>7.5</td>
<td>25g: add 20 mL salt water</td>
<td>2.88x, 80% Ethanol + 0.12x, 7% Acetic Acid</td>
<td>1 min – shaker or 2 min – by hand</td>
<td>Filter; mix well</td>
<td>400 µL</td>
<td>100 µL</td>
<td>4 min</td>
</tr>
</tbody>
</table>

TIPS!

**Get Complete Extraction**
- Fully wet samples before shaking
- Assure liquid is moving forcefully though the sample while shaking

**For Best Performance**
- Pipette up and down while mixing
- Do not reuse diluted samples
- Read strips promptly after run time

**Avoid Contamination**
- Use a new reaction vial per test
- Keep DB5 capped, when possible
- Use new pipette tips each step
SET F Procedures (Incubator required): Additional Matrices

**Matrices:**
- Peanut Seed (high sensitivity)

Review Sample Preparation on page 3 for grinding consistency and notes

### Sample Extraction

Prepare a bulk salt water solution: (a) Add 29.4g of table salt (non-iodized) per 100 mL of bottled water. (b) Mix well, until salt is in solution.

<table>
<thead>
<tr>
<th>25g Samples</th>
<th>50g Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Create Slurry</strong></td>
<td><strong>Create Slurry</strong></td>
</tr>
<tr>
<td>1. Add 10 mL salt water to sample</td>
<td>1. Add 20 mL salt water to sample</td>
</tr>
<tr>
<td>2. Mix well, stir slowly</td>
<td>2. Mix well, stir slowly</td>
</tr>
<tr>
<td><strong>Add Solvent</strong></td>
<td><strong>Add Solvent</strong></td>
</tr>
<tr>
<td>3. Add 50 mL 80% Ethanol</td>
<td>3. Add 100 mL 80% Ethanol</td>
</tr>
<tr>
<td>4. Make sure entire sample is wetted</td>
<td>4. Make sure entire sample is wetted</td>
</tr>
</tbody>
</table>

The above extraction protocol provides instruction for 25g and 50g samples. To customize sample size, keep salt water slurry and Ethanol ratios the same.

**Turn on incubator** and set the temperature to 22°C, let equilibrate for at least 10 minutes.

**Shake:** choose mechanical shaker or hand shaking

| Shaker Table: mix at highest speed for 1 minute | By Hand: shake vigorously for 2 minutes |

**Clarity Extract:**
Pour through an approved coffee filter (e.g. ACC-083). Mix the clarified extract well before testing.

**Combine Buffer and Extract, then Run Test Strips**
1. **Consult TABLE F** to determine DB5 and extract volume
2. Add DB5 to the 12X75mm reaction tube (discard tip)
3. Add clarified extract to the reaction tube
4. Mix thoroughly with extract pipette tip (discard tip)
5. Insert tube into incubator
6. *Wait 2 minutes (equilibration time)
7. Add test strip to tube, arrows down
8. Wait 4 minutes (run time)
9. Immediately cut strips at the top of the arrow tape (discard bottom pads)
10. Insert strip, barcode face down, into QuickScan reader
11. When prompted, select Matrix Group for the matrix being tested

**TIPS!**

- Get Complete Extraction
  - Fully wet samples before shaking
  - Assure liquid is moving forcefully though the sample while shaking

**For Best Performance**
- Pipette up and down while mixing
- Do not reuse diluted samples
- Read strips promptly after run time

**Avoid Contamination**
- Use a new reaction tube per test
- Keep DB5 capped, when possible
- Use new pipette tips each step

**TABLE F: High Sens. Peanut Seed Matrix Summary Guide**

<table>
<thead>
<tr>
<th>Matrix</th>
<th>Matrix Group</th>
<th>LOD (ppb)</th>
<th>Slurry</th>
<th>Extractant</th>
<th>Shake</th>
<th>Clarify</th>
<th>DB5 Volume</th>
<th>Extract Volume</th>
<th>Add tube to Incubator</th>
<th>Run Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>High Sens. Peanut Seed</td>
<td>MG17</td>
<td>2.5</td>
<td>25g: add 10 mL salt water 50g: add 20 mL salt water</td>
<td>2x, 80% Ethanol</td>
<td>1 min – shaker or 2 min – by hand</td>
<td>Filter; mix well</td>
<td>300 µL</td>
<td>200 µL</td>
<td>Acclimate tube for 2 min*</td>
<td>4 min</td>
</tr>
</tbody>
</table>

*The tube acclimation step is only required if the temperature of the testing environment is unknown or outside of 20-24°C (68-75°F)
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