ENVIROLOGIX

QuickTox™ Kit for QuickScan Fumonisin

Catalog Number AQ 111 BG *Part #10421*

Highlights:

- *Quantitative results in only 5 minutes*
- *Read strips wet – no drying necessary*
- *Simple protocol*
- *No incubation equipment needed*
- *Common Extraction with QuickTox Kit for Aflatoxin*

Contents of Kit:

- *50 QuickTox Strips packed in a moisture-resistant canister*
- *100 vials*
- *100 pipette tips*
- *DB2 Buffer*
- *Available in 50-strip individual kit format or bulk packaging*

Items Not Provided:

- *Orbital/rotary shaker*
- *Plastic sample cups with lids**
- *Solvent (50% ethanol)**
- *20 mesh screen*
- *Graduated cylinder**
- *Pipette to deliver 100 µL**
- *Pipette to deliver 1 mL (for dilution of high samples)*
- *Timer*
- *Scissors*
- *QuickScan System**

**Available as accessories – see list on Page 4*

Correct 20 mesh grind for corn

Intended Use

This EnviroLogix QuickTox Kit for QuickScan Fumonisin is designed to quickly extract and screen corn and sorghum for the presence of fumonisin residues. The QuickTox Kit is designed to provide quantitative results in corn grain and bulk sorghum for fumonisin residues ranging from 0.20 ppm to 6.0 ppm in the standard assay, and to 20 ppm with additional dilution. For quantification of fumonisin levels above 20 ppm, please contact Tech Service.

How the Test Works

A composite sample is first collected, then extracted to solubilize any fumonisin present. Each sample should be ground to a fineness of 20 mesh and extracted with diluted solvent. This extract is further diluted for testing with the QuickTox Kit.

Each QuickTox Strip has an absorbent pad at each end. The protective tape with the arrow indicates which end of the strip to insert into the reaction vial. The sample extract travels up the membrane strip and is absorbed into the larger pad at the top of the strip. At five minutes, the strip is cut off 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.

Preparation of the Sample

Please note: sample extract should be tested shortly after dilution with Buffer (Step 8). Make sure strips and Buffer are at room temperature and ready for use before the dilution step.

Determine size of sample

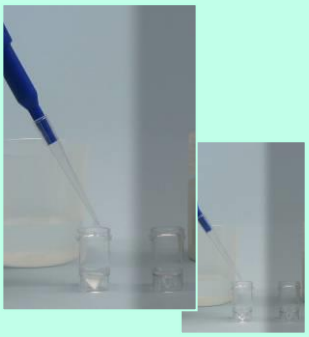
1. Collect a composite sample according to your own sampling plan or USDA/GIPSA guidelines. Consult USDA/GIPSA reference documents such as **www.gipsa.usda.gov/publications/fgis/handbooks/gihbk1_insphb.html** to help design a plan that fits your needs.
2. Grind samples using a grinder or mill which provides a sample that passes through a 20 mesh sieve. For example, if using a Bunn grinder, the Turkish setting yields the correct grind. Mix ground material thoroughly before sub-sampling.

Extract sample

3. Weigh 20 to 50 grams of milled sample into a disposable sample cup with lid or other suitable container and add two volumes of 50% ethanol (2 mL per gram of sample, i.e. 20 grams, add 40 mL). To purchase or prepare a 50% ethanol solvent, see Precautions & Notes. Keep ethanol covered and work in a draft-free area to minimize evaporation during testing.
4. Cap sample cup tightly and place on shaker for 1 minute at highest speed, or shake vigorously by hand for 1½ to 2 minutes. Samples that are not thoroughly mixed may adversely affect test results due to incomplete extraction.

Measure solvent, add to ground sample

Shake mechanically or by hand



Add 50% ethanol then the sample extract to the first vial (the dilution vial) and mix. Discard pipette tip.

With a new pipette tip, add DB2 Buffer to the second vial (the reaction vial) for testing

Then transfer diluted sample from dilution vial to the reaction vial, mix well with pipette tip

5. Extract will immediately begin to separate into layers (a more finely ground beginning sample may take a few minutes to settle). The top (yellowish) layer containing the fumonisin residues will be used in testing.

Dispense liquids (use 2 vials and 2 pipette tips for each sample being tested: the first set for dilution, the second set for testing):

Dilution (vial #1):

6. Using a calibrated pipette with a **new tip**, place 100 microliters (100 µL) 50% ethanol into the (first) dilution vial.
7. Using the same tip, remove 100 µL from the top (yellowish) layer of extract from the sample cup, avoiding particulates. Add extract to dilution vial containing the ethanol and mix well with pipette by stirring or drawing liquids up and down in the pipette tip. Then discard that pipette tip.

Testing (vial #2):

8. With **a new** pipette tip**,** add 100 µL of DB2 Buffer to the (second) reaction vial, used for testing**.** Take care not to contaminate Buffer—use a new tip for each test, pipette Buffer before pipetting the diluted extract, and keep Buffer bottle covered when not in use.
9. Using the same tip, transfer 100 µL of the well mixed **diluted sample extract** from the dilution/first vial into the reaction/second vial containing the DB2 Buffer, and mix well by stirring or drawing the liquids up and down in the pipette tip until the mixture is uniformly light yellow.

NOTES: Samples that are not thoroughly mixed and/or accurately pipetted will adversely affect test results. Do not reuse diluted samples. Use two new vials for each sample. Use two pipette tips for each sample (one for solvent and extract in the dilution vial, and one for DB2 Buffer and diluted extract in the reaction vial).

For testing samples at levels greater than 6 ppm (up to 20 ppm):

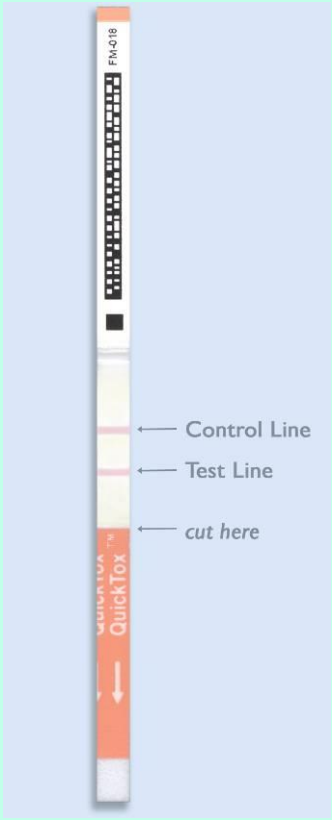
If after running and reading the test, the initial result is greater than 6 ppm ("> 6.0 ppm" on QuickScan), and further knowledge about the level of contamination is desired, higher levels can be estimated by further dilution of the sample extract.

1. In a new vial (Dilution: vial #1) combine 100 µL (50% ethanol) + 100 µL extract from the top layer of the original extraction (measure carefully). Add 1 mL 50% ethanol into the same vial. **Mix well**.
2. Using a calibrated pipette with a **new tip**, place 100 µL DB2 Buffer into a second vial (Testing: vial #2).
3. With the **same** pipette tip, add 100 µL of the newly diluted extract to the reaction vial containing DB2 Buffer. **Mix thoroughly.** Because the extract is further diluted, its addition to the buffer may not turn the mixture yellow.
4. Follow the instructions under How to Run. Choose 1:6 under the dilution tab on QuickScan Results Screen—the System will calculate and record the fumonisin level in diluted samples.

How to Run the QuickTox Strip Test

1. Allow refrigerated canisters to come to room temperature before opening. Remove the QuickTox Strips to be used. Avoid bending the strips. Reseal the canister immediately.

Place strip in reaction vial. Wait 5 minutes for results.



Cut strip and place in QuickScan reader immediately —no drying step!

2. Place the strip into the reaction vial containing the Buffer and diluted sample extract (Testing: vial #2). The arrow tape on the end of the strip should point into the reaction vial.
3. The liquid will travel up the strip (flow may not be visible immediately—this is expected and normal).
4. Allow the strip to develop for 5 minutes. Immediately cut off and discard the bottom section of the strip covered by the arrow tape, and insert the strip into the QuickScan reader for quantitation.

Use of the QuickScan System

Detailed instructions for use of the QuickScan System are supplied with each unit, and can also be found at envirologix.com/quickscan. The lot-specific Multi-Matrix Barcode Card (MMBC) must be scanned into the system prior to testing. In summary, a strip is inserted into the reader and the strips are read by touching or clicking on the "Read Test" area of the screen. Results are then recorded in an electronic worksheet, allowing each user to report and track data easily.

Results are reported in the range of 0.20 to 6.0 ppm. Results less than 0.20 ppm are reported as "<LOD" (less than Limit of Detection) and results greater than 6.0 ppm are reported as "> 6.0 ppm."

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. 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 levels stated.

- Aflatoxin B1 – 2000 ppb
- DON (deoxynivalenol) – 2000 ppm
- Ochratoxin A – 2000 ppm
- Zearalenone – 2000 ppm

Precautions and Notes

- This product is currently not applicable for use in testing any other crops.
- Pipettes lose calibration accuracy over time. Calibrate or replace pipettes at least annually.
- This assay is calibrated against reference samples supplied by Trilogy Analytical Laboratory, Washington, MO, and other vendors and associated HPLC data.
- 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 protocol provided in the kit. Deviation from this protocol may invalidate the results of the test. Proper and thorough mixing, along with 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 seed lot 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 or protein in question.

- Strips must be read wet promptly at five minutes.
- Protect all components from hot or cold extremes of temperature when not in use. Do not leave in direct sunlight or in vehicle.
- For convenience, accessories and solvent can be ordered from EnviroLogix (see list, below). Purchase 50% ethanol, or prepare using 100% ethanol as follows: 50% Ethanol Preparation Instructions: For 100 mL, measure 50 mL 100% ethanol [reagent grade or better]; pour into suitable container with cap. Add 50 mL deionized or distilled water. Cap tightly and shake to mix. Use care when uncapping.
- **IMPORTANT:** Ethanol is flammable and toxic. 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. Observe any applicable regulations when disposing of samples and kit reagents.
- Any reusable labware contacting fumonisin (example: glass collection jars) should be washed thoroughly in warm, soapy water, and rinsed completely before reuse.

Accessories:

Available through EnviroLogix:	*Catalog No.*	*Part #*
▪ QuickScan™ System	ACC 331	12721
▪ Sample cups with lids (500/case) *for samples up to 30 g; larger samples require different mixing vessels*	ACC 012-CS	10167
▪ Graduated cylinder (100 mL)	ACC 068	11207
▪ MiniPet pipette 100 µL (one/location free)	ACC 041	11202
▪ 50% Ethanol	ACC E26902-1X4 (1 bottle, 4L)	11156

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License

EnviroLogix has developed this kit using proprietary reagents.