

# QuickStix<sup>™</sup> Combo Kit for Cry1Ac/Cry2A/LibertyLink<sup>®</sup>/bar/Cotton Seed

#### Highlights:

- Recognizes Cry1Ac, Cry2A and LibertyLink (bar), and will not cross-react with PAT/pat protein
- Results in 10 minutes or less

#### Contents of Kit:

- 100 QuickStix Combo Strips packed in two moistureresistant canisters
- 100 Extraction Tubes
- EB2 Extraction Buffer

#### Items Not Provided:

• Repeating pipetter or other means of dispensing 0.5 mL

Contact EnviroLogix to order bulk-packaged combs. Bulk kits include 20x Buffer Concentrate instead of EB2. To prepare, mix 50 mL Buffer Concentrate with 950 mL of distilled or deionized water. Store refrigerated when not in use; warm to room temperature before using.



Crush single seed



Catalog Number AS 047 ST

## Intended Use

The EnviroLogix QuickStix Combo Kit for Cry1Ac/Cry2A/LibertyLink is designed to detect the presence of these traits at the levels typically expressed in genetically modified cotton seed. The combo strips will recognize Cry1Ac, Cry2A and PAT/bar in separate regions of the same strip. It is the only lateral flow strip capable of detecting only the PAT/bar protein; it will not cross-react with the PAT/pat protein.

## How the Test Works

Cotton plants and seeds that have been genetically modified with stacked genes express transgenic proteins in their seed. To detect these proteins with the EnviroLogix QuickStix Combo Strip, the sample must be extracted and the proteins solubilized in the Extraction Buffer provided.

Each Combo Strip has an absorbent pad at each end. The protective tape with the arrow indicates which end of the strip to insert into the sample. The sample travels up the membrane strip and is absorbed into the larger pad at the top of the strip. The portion of the strip between the protective tape and the absorbent pad at the top of the strip is used to view the reactions as described under "Interpreting the Results".

## Sample Preparation

**Note:** If Extraction Buffer has been refrigerated, allow it to warm up to room temperature before preparing samples.

#### To extract cotton seed:

- 1. Crush a single cotton seed (Suggestion: use pliers with seed in microcentrifuge tube or resealable plastic bag). Transfer to an extraction tube marked with sample identification. Note: Complete crushing of seed improves extraction efficiency and test performance.
- 2. Add 0.5 mL Extraction Buffer.
- 3. Close the tube cap securely. Shake the tube vigorously for 20 to 30 seconds, using an **up-and-down motion**, ensuring that the crushed seed and buffer are **well** mixed. Allow the solid material to settle to the bottom of the tube. The extract takes on a yellow to brown opaque color when the samples are prepared properly.
- 4. Use caution to prevent sample-to-sample cross-contamination with plant tissue, fluids, crushing equipment (*pliers*) or disposables. Be sure to use a new tube for each sample tested.

# How to Run the QuickStix Strip Test

- Allow refrigerated canisters to come to room temperature before opening. Remove the QuickStix Strips to be used. Avoid bending the strips. Reseal the canister immediately.
- 2. Place the strip into the cotton seed extract. The sample will travel up the strip. Use a rack to support multiple tubes if needed.

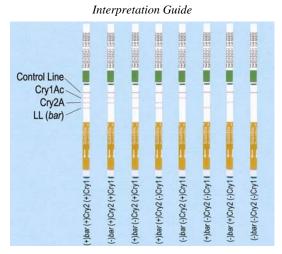
- 3. Allow the strip to develop for 10 minutes before making final assay interpretations. Positive sample results may become obvious much more quickly.
- 4. To retain the strip, cut off and discard the bottom section of the strip covered by the arrow tape.

# Interpreting the Results

Development of the Control Line within 10 minutes indicates that the strip has functioned properly. Any strip that does not develop a Control Line should be discarded and the sample re-tested using another strip.

If the extract is from a negative sample, the strip will show only a single line, the Control Line, about 6 mm below the top pad.

If the extract is from a sample containing Cry1Ac proteins, Cry2A proteins, and/or PAT/bar proteins, the appropriate test line(s) will appear. A Test Line for extracts containing Cry1Ac protein will appear about 4 mm below the Control Line. A Test Line for extracts containing Cry2A protein will appear about 8 mm below the Control Line and approximately 4



mm below the Cry1Ac Test Line. A Test Line for extracts containing PAT/bar protein will appear about 12 mm below the Control Line and approximately 4 mm below the Cry2A Test Line.

Please note that for ease of interpretation, the labels on each strip list the proteins in the order that their Test Lines appear, bottom (arrow end) to top (lot number end).

# Kit Storage

This Kit can be stored at room temperature, or refrigerated for a longer shelf life. Please note the shelf life on the kit label for each storage temperature. The kit may be used in field applications; however, prolonged exposure to high temperatures may adversely affect the test results. Do not open the desiccated canister until ready to use the test strips.

## **Precautions and Notes**

- This kit is designed for screening for presence or absence only and is not meant to be quantitative.
- This product is currently not applicable for use in any other crop than cotton.
- As with all tests, it is recommended that results be confirmed by an alternate method when necessary.
- The assay has been optimized to be used with the protocol and with the buffer provided in the kit. Deviation from this protocol may invalidate the results of the test.

Any clearly discernable pink Test Line is considered positive



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- A negative result with this kit does not mean that the sampled tissue has not been otherwise genetically modified.
- A strong positive result may safely be interpreted in as little as 2 minutes after sample addition. It is not safe, however, to conclude that a sample is negative before a full 10 minutes has elapsed.
- Protect all components from hot or cold extremes of temperature when not in use. Do not leave in direct sunlight or in vehicle.

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