

Highlights:

- Results in 5 minutes or less
- Available in 100-strip individual kit format or bulk packaging

Contents of Kit:

- 100 QuickStix Strips packed in two moisture-resistant canisters
- 100 Disposable Tissue Extractors, each consisting of a tube with punch cap and pestle (optional item with bulk packaging)
- 1 dropper bottle for dispensing tap water



Obtain Leaf Tissue



Grind Tissue

Catalog Number AS 010 LS

Intended Use

The EnviroLogix QuickStix Kit for Roundup Ready® Plant Tissue screens for the presence of CP4 EPSPS protein in **Corn Leaf, Corn Seed, and Soy Leaf** tissues. It is not currently validated for any other crops. The QuickStix will not detect GA21 Roundup Ready.

If you wish to test soybean or corn bulk grain, order Catalog Number AS 010 BG. If you are interested in testing canola leaf and seed, order Catalog Number AS 017 LS.

How the Test Works

Crops that have been genetically modified with Roundup Ready® express CP4 EPSPS protein in their tissue. To detect the CP4 EPSPS protein with the EnviroLogix QuickStix Strip, tissue samples must be extracted and the protein solubilized in tap water.

Each QuickStix Strip has an absorbent pad at each end. The protective tape with the arrow indicates which end of the strip to insert into the extraction tube. 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

Prior to beginning, fill the dropper bottle provided with water.

To extract corn leaf tissue:

1. Sandwich a section of leaf tissue between the cap and body of the Disposable Tissue Extractor tube. Snap **one** circular tissue punch by closing the cap. Push the leaf punch down into the tapered bottom of the tube with the pestle. Write the sample identification on the tube with a waterproof marker.
2. Insert the pestle into the tube and grind the tissue by rotating the pestle against the sides of the tube with twisting motions. Continue this process for 20 to 30 seconds, or until the leaf tissue is well ground.
3. Holding the dropper bottle vertically, carefully add 10 drops of water into each tube containing corn leaf tissue. If using a pipette, add 500 µL of water.
4. Repeat the grinding step to mix tissue with water. Dispose of the pestle. Do not re-use pestles on more than one sample.

To extract corn seed:

1. Crush a single corn seed (*Suggestion: use pliers with seed in microcentrifuge tube or resealable plastic bag*). Transfer to a tissue extraction tube marked with sample identification. Note: Complete crushing of seed improves extraction efficiency and test performance.
2. Holding the dropper bottle vertically, add 20 drops of water into the tube. If using a pipette, add 1 mL of water.
3. Close the tube cap securely. Shake the tube vigorously for 20 to 30 seconds. Allow the solid material to settle to the bottom of the tube.



Crush Seeds



Extract Seed Sample



Insert QuickStix Strip



Any clearly discernable pink Test Line is considered positive

- Repeat the protocol for each sample to be tested, using a new tube for each. Use caution to prevent sample-to-sample cross-contamination with plant tissue, fluids, crushing equipment (*pliers*) or disposables.

To extract soy leaf tissue:

Note: Tissue from leaf sets 1-4 or cotyledons should be selected when testing plants younger than 36 days. Since soy leaves are high expressers of CP4 EPSPS protein take care to avoid cross-contamination when sampling the leaf tissue. Plant tissue older than 36 days should be sampled from cotyledon or leaf set 1 or 2 only.

- Sandwich a section of tissue between the cap and body of the Disposable Tissue Extractor tube.
 - For leaf tissue, snap **one** circular tissue punch by closing the cap. The soy leaves lowest on each plant (first or second set above the cotyledons), should be used.
 - For cotyledons, snap $\frac{1}{2}$ circular tissue punch by placing tissue halfway across the tube before closing the cap.
- Push the tissue punch down into the tapered bottom of the tube with the pestle. Write the sample identification on the tube with a waterproof marker. **Do not grind the tissue.** Do not re-use pestles on more than one sample.
- Holding the dropper bottle vertically, carefully add 10 drops of water into each tube containing unground leaf or cotyledon tissue. If using a pipette, add 500 μ L of water.
- Close the tube cap securely. Shake the tube vigorously for 20 – 30 seconds. Allow the solid material to settle to the bottom of the tube.

How to Run the QuickStix Strip Test

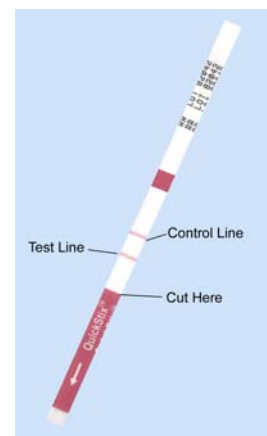
- Allow refrigerated canisters to come to room temperature before opening. Remove the QuickStix Strips to be used. Avoid bending the strips. Reseal the canister immediately.
- Place the strip into the extraction tube. The sample will travel up the strip. Use a rack to support multiple tubes if needed.
- Allow the strip to develop for 5 minutes before making final assay interpretations. Positive sample results may become obvious much more quickly.
- If you wish 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 5 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 sample extract contains the CP4 EPSPS protein, a second line (Test Line) will develop on the membrane strip between the Control Line and the protective arrow tape. The results should be interpreted as positive for CP4 EPSPS protein expression.

If no Test Line is observed after 5 minutes, the results should be interpreted as negative. A negative result means the sample contains less CP4 EPSPS than is typically expressed in the tissues of Roundup Ready plants.





Kit Storage

This QuickStix Kit can be stored at room temperature, or refrigerated for a longer shelf life. Note the shelf life on the kit box 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.

Cross-reactivity

The following materials have been tested with this kit and have been found to cause no false positive results:

- Proteins: Cry1Ab, Cry1Ac, Cry1C, Cry1F, Cry2Aa, Cry2Ab, Cry3Bb, Cry9C, PAT/*pat*, PAT/*bar*, and cyanamide hydratase at concentrations at least 4 times higher than reported in commercial crops.
- Other GMO Event Corns: YieldGard[®] Corn Borer (Bt11, MON810), Knockout[®]/NatureGard[®] (Bt176), LibertyLink[®] (T25), Starlink[®], Roundup Ready (Event GA21), YieldGard[®] Rootworm Corn, and Herculex[™] I corn leaf and seed.
- Other crops: Leaf tissue from conventional varieties of barley, canola, corn, cotton, rice, sorghum, soybean, sugarbeet, and wheat at 100% of sample.

Precautions and Notes

- This kit is designed for screening for presence or absence only and is not meant to be quantitative.
- 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 provided in the kit. Deviation from this protocol may invalidate the results of the test.
- The results generated through the proper use of this kit 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.
- 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 5 minutes has elapsed. A weakly positive sample may require the full 5 minutes for a distinct Test Line to appear.
- Protect all components from hot or cold extremes of temperature when not in use. Do not leave in direct sunlight or in vehicle.



**For Technical Support
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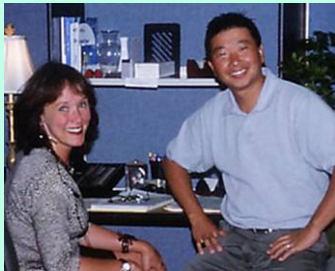
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