

QuickStix™ Field Test Kit for PAT/*bar* Cotton Leaf

Catalog Number AS 013 LTF

Intended Use

The EnviroLogix QuickStix Kit for PAT/*bar* Plant Tissue screens for the presence of PAT/*bar* protein in **cotton leaf** tissues. The QuickStix will not detect PAT/*pat* protein; it is specific to PAT/*bar* only.

How the Test Works

This test is for use on cotton leaf tissue that has been genetically modified to express PAT/*bar* protein. To detect the PAT/*bar* protein with the EnviroLogix QuickStix Strip, leaf samples must be extracted and the protein solubilized in the buffer provided.

Each QuickStix Strip has an absorbent pad at both ends. 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".

Please refer to the inside of the kit box for a pictorial guide to running the test.

Sample Preparation

To extract cotton leaf tissue:

1. Sandwich a section of leaf tissue between the cap and body of the Disposable Tissue Extractor tube. Snap **one or two** circular leaf punches 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. Holding the dropper bottle vertically, carefully add 10 drops of buffer into each tube containing cotton leaf tissue. If using a pipette, add 500 µL of buffer.
3. 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.
4. Dispose of the pestle. Do not re-use pestles on more than one sample.

How to Run the QuickStix Strip Test

1. 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 extraction tube. 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 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 PAT/*bar* 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 PAT/*bar* protein expression.

If no Test Line is observed after 10 minutes, the results should be interpreted as negative. A negative result means the sample contains less PAT/*bar* than is typically expressed in the tissues of modified cotton 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.

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 10 minutes has elapsed. A weakly positive sample may require the full 10 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. A small portable cooler is recommended for field testing applications to protect the kit from extreme temperatures.

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