

QuickStix[™] Kit for Roundup Ready[®] Alfalfa Bulk Seed

Catalog Number AS 045 BG

Contents of Kit:

- 100 QuickStix Strips packed in two moisture-resistant canisters
- 100 Extract Filter units
- 100 10 mL graduated Tubes with Caps

Items Not Provided:

- Proctor-Silex E160B Coffee Grinder
- Weighing paper, or equivalent







Intended Use

This EnviroLogix QuickStix Kit for Roundup Ready in Alfalfa Bulk Seed is designed to extract and detect the presence of CP4 EPSPS protein in ground alfalfa seed at levels of 0.167% or above, dependent upon the expression level of the Roundup Ready plant. One Roundup Ready alfalfa seed in a total of 600 alfalfa seeds equals 0.167%.

The test is intended to give the producer, purchaser, or exporter a screening method for identification of the presence of Roundup Ready alfalfa seed in a bulk seed lot at the level indicated above. The results of each sample tested are only as representative as the sample is of the entire lot, so adherence to proper principles of sampling is key.

How the Test Works

In order to test for the Roundup Ready protein expressed by Roundup Ready alfalfa seeds, the sample must first be finely ground and then extracted with water. Seed extracts are prepared for testing using a prescribed method (see Sample Preparation section). Testing extracts from seed preparations that have not been properly ground and extracted can lead to incorrect results.

Each QuickStix Strip has an absorbent pad at each end. The protective tape with arrows indicates the end of the strip to insert into the sample extract. 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 Collection

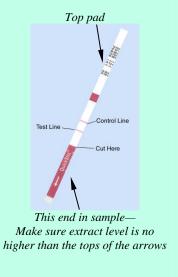
It is very important that the sample tested is representative of the entire lot of seeds. One sample of 600 seeds (or approximately 1.7 grams) for each seed lot is required to run the test.

Sample Preparation

- 1. Weigh a 600 seed sample (equal to approximately 1.7 grams) onto weighing paper using a scale.
- 2. Place the sample in a clean coffee grinder, making sure that <u>all</u> seeds are added. The recommended coffee grinder is Proctor-Silex Fresh GrindTM Model E160B.
- 3. Grind the sample in the grinder for five pulses, lasting 5 seconds each. After the last grind, examine the ground seeds to be sure all are broken.
- 4. Carefully transfer the ground seeds back to weighing paper, and then place in a 10 mL tube.
- 5. Add 4 mL of water to the tube and secure cap on tube.
- 6. Immediately shake tube containing ground seeds and water for 15 seconds only.







<u>Note</u>: The alfalfa seeds absorb water very quickly and must be processed *promptly* to ensure optimal performance.

7. After shaking, immediately insert the filter into the tube and push it down until the red rubber head reaches the 5 mL line on the graduated tube. Approximately 1/4" of cloudy extract should be visible in the filter unit.

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 filter unit containing extract. The sample will travel up the strip.
- 3. Allow the strip to develop for 15 minutes before making final assay interpretations.
- 4. To retain the strip, remove strip from filter unit using tweezers, then cut off and discard the bottom section of the strip covered by the arrow tape.

NOTE: Use extreme caution to prevent sample-to-sample cross-contamination with seeds, fluids, or disposables. Do not re-use filters.

Interpreting the Results

Development of the Control Line within 15 minutes indicates that the strip has functioned properly. Any strip that does not develop a Control Line should be discarded, and the sample retested using another strip.

If the extract is from a sample containing at least 0.167% Roundup Ready alfalfa (1 seed in 600), a second line (Test Line) will develop on the membrane strip between the Control Line and the protective tape. The results should be interpreted as positive for CP4 EPSPS protein, indicating the presence of a Roundup Ready alfalfa seed.

If the extract is from a negative sample, the strip will only develop the Control Line.



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 meant to be a qualitative test.
- The strips will detect Roundup Ready protein in sample extracts prepared following the specified extraction procedure at levels of 0.167% or more and is dependent upon the expression level of the Roundup Ready alfalfa plant.
- This product is currently not applicable for use in any other crop.
- Testing extracts from seed preparations that have not been properly ground and extracted can lead to incorrect results.
- As with all tests, it is recommended that results be confirmed by an alternate method when necessary.
- The assay has been optimized 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 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.
- It is not safe to conclude that a sample is negative before a full 15 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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