## Rapid lateral flow strip (LFS) for detection of PAT/pat in maize / soybean seed or leaf Cat No. AID 029

## Intended use

PAT/pat (Liberty link) lateral flow strips (LFS) test kit is intended to be used for the qualitative detection of PAT/pat protein in individual soybean and corn leaf or seed samples.

The total incubation time of the assay is 5 minutes.

## **Principle of the test**

An antibody specific to the PAT protein molecule is immobilized on the test line area of nitrocellulose membrane. Second antibody specific for PAT molecule is conjugated with colloidal gold and incorporated in the sample area of lateral flow strip. Anti mouse IgG is immobilized on the control line area of the LFS.

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When the LFS is placed in the sample extract, the PAT protein present in the sample extracts binds to the antibody labelled with gold nano particles and the complex moves upward by capillary action. The complex then binds to the antibody coated on the test line resulting in pink/purple color test line. As the complex moves further up, it binds to the control line resulting in pink/purple color control line. In absence of PAT, the test line does not appear as no complex binds to the test line while control line turns pink/purple color indicating validity of test protocol.

#### **Cross-reactivity**

The PAT/Pat LFS kit does not cross react with CP4EPSPS (RoundUp Ready\*\*), Cry1F, Cry1Ac, eCry3.1Ab (AgriSure Duracade), Cry3Bb, Cry1Ab, Cry2A, Vip3A, Cry34Ab1 or Cry35Ab1.

#### **Contents of the kit: Kit is sufficient for 100 tests**

PAT/pat LFS st	rips 50 strips pe	<mark>er canister. T</mark> wo canisters per kit	
Extraction buffe	er Two vials o	Two vials of 50ml each. Ready to use	
Pack insert	One		
Dropper	One		

#### Material and equipment required but not provided

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- 1.5 ml Microfuge tubes with cap
- 48 well seed crushing plate \* Pair of scissors
- Pliers •••

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48 prong seed crusher

Timer

Hammer

Marking pen and Paper Towels <a> Pestles</a> \*

#### **Precautions**

The PAT LFS kit is intended for in vitro use only. The reagents contain sodium azide as preservative. Prevent direct skin and eye contact with kit components. Seek medical attention in case of accidental ingestion of kit components.

#### Storage of the kit

The kit should be stored at 2 - 8° C. The unopened kit is stable till the expiry date printed on the kit label. The cap of the canister should be closed firmly after removing the required strips. Exposure to moisture is likely to affect the performance of the test strips.

The kit should not be frozen.



# **Technologies**

Extracting protein by crushing with pestle



#### **Sample preparation**

#### Extraction of seed tissue:

Crush single seed and transfer it to 1.5 ml microfuge tube. Add 1.0 ml of extraction buffer. Mix well and wait for 5 minutes.

#### **Extraction of leaf tissue:**

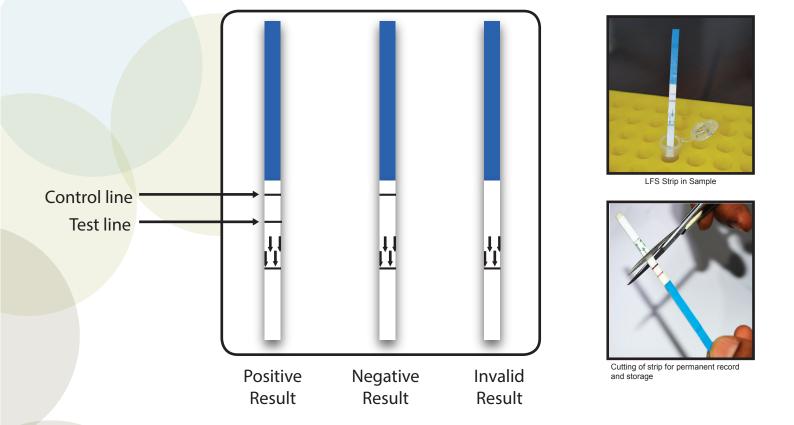
Take two leaf punch (approximate weight 20 mg) and transfer it to 1.5 ml microfuge tube. Grind the leaf tissue with pestle until the leaf tissue is well ground. Add 0.5 ml extraction buffer. Mix well and wait for 5 minutes.

Use new pestle for each leaf sample to avoid cross contamination.

Note: Test the sample within 30 minutes after extraction.

#### Assay Procedure

- 1. Allow canister to come to room temperature before opening it to remove the desired number of strips.
- 2. Insert one strip in each sample. Part of the strip showing arrow should be dipped in sample extract. Allow the test strip to remain in the microfuge tube in vertical position for 5 minutes.
- 3. Remove the strips and observe the results. Positive sample results may appear much earlier than 5 minutes.
- 4. For permanent storage of strips, cut off the bottom section of the strip covered with arrow using pair of scissors.



## **Interpretation of LFS results**

Read the strip in 5 minutes.

Two line indicates positive test result while single line indicates negative results. Absence of control line in 5 minutes indicates invalid test.

The appearance of faint test line after 5 minutes should not be necessarily interpreted as positive test.

#### <u>Notes</u>

- The procedure instructions should be strictly followed to get correct results. Change in procedure may lead to wrong results.
- This kit has been optimized for individual Soybean/Corn seed and leaf.
- Cross contamination between different microfuge tubes will lead to wrong results
- This kit is meant for screening for presence or absence of PAT/pat in individual Soybean/Corn seed and leaf.
- When in doubt, please confirm results with an alternate method.

#### WARRANTY

Eurofins Amar Immunodiagnostics Pvt. Ltd. warrants that the products sold hereunder ("the Product") are defectfree in material and workmanship, provided they are used in accordance with the prescribed instructions before the expiry of the products as printed on the product label.

The customer should notify Eurofins Amar Immunodiagnostics in writing of Warranty defects during the warranty period, including an offer by the customer to return the Products to Eurofins Amar Immunodiagnostics for evaluation. Eurofins Amar Immunodiagnostics will repair or replace, at its sole option, any product or part thereof that proves defective in materials or workmanship within the warranty period.

This warranty also does not apply to Products to which changes or modifications have been made or attempted by persons other than pursuant to written authorization by Eurofins Amar Immunodiagnostics

#### THIS WARRANTY IS EXCLUSIVE

The sole and exclusive obligation of Eurofins Amar Immunodiagnostics shall be to repair or replace the defective Products in the manner and for the period provided above.

Eurofins Amar Immunodiagnostics shall not have any other obligation or liability, whatsoever it may be, with respect to the Products or any part thereof. Under no circumstances, whatsoever the circumstances may be, shall Eurofins Amar Immunodiagnostics be liable for incidental, special, or consequential damages.

If any part of this Warranty is determined to be void or illegal, the remainder shall remain in full force and effect.

\*Parafilm is a registered trademark of American Can Corporation (now Pechinney Plastic Packaging).

\*\*Bollgard & Roundup Ready are registered trademarks of the Monsanto Company.

\*\*\*Liberty Link is a register trademark of Bayer Crop Science.

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