Pro-Detect™ Rapid GST Assay Kit

Catalog Numbers A38502

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WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from **thermofisher.com/support**.

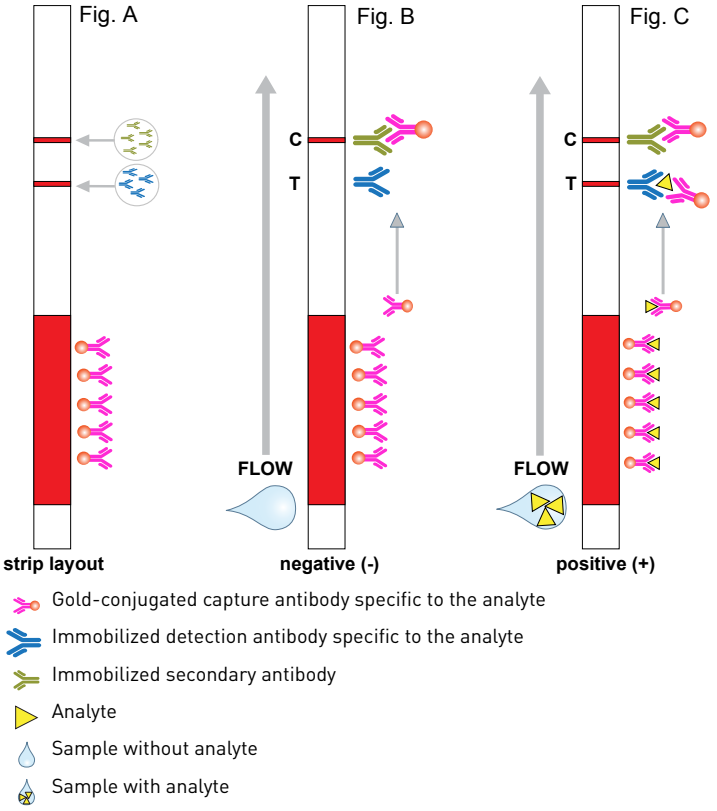
Contents and storage

Product	Cat. No.	Contents	Storage
Pro-Detect™ Rapid GST Assay Kit	A38502	Pro-Detect™ Rapid GST Assay Strips, 10 strips Pro-Detect™ Rapid Assay Dilution Buffer, 15 mL	Store at 4°C. Do not freeze. After opening, store unused strips in the enclosed container containing desiccant.

Product description

The Pro-Detect™ Rapid GST Assay Kit is a 10-minute dipstick lateral-flow assay that detects GST-tagged proteins from cell culture media or lysate and purified protein preparations. The assay is performed by simply applying the lateral flow strip into the sample of properly diluted tissue culture supernatant or lysate and visualizing via red bands at the test and control lines.

Embedded in the conjugate pad at the bottom of the strip, the gold-conjugated capture antibody is specific to GST so it binds to the GST tag bound to the end of the protein. Additionally, a detection antibody specific to the GST end of the protein is embedded at the test line, while a control antibody is embedded in a control line at the top of the strip (Fig. A). Upon application, the capture antibodies, bound to any present GST protein tag, travel the length of the membrane and are resolved on the test and control lines, displayed visually by red bands. If no test band displays, then the sample is negative (Fig. B), while the display of both the test line and the control line indicates the presence of tagged protein in the sample (Fig. C).



Material preparation

The recommended working range of the GST lateral flow assay is 0.1 µg/mL to 10 µg/mL. The lateral flow assay will be able to detect outside of this range, however intensity of the bands will be lower at concentrations higher and lower than the working range. It is not recommended to use at concentrations less than 0.01 µg/mL.

Note: Proper sample dilution is essential for optimal results. Concentration ranges are based upon the concentration of the tagged protein of interest in the sample.

Sample dilution

If the concentration of the GST-tagged protein in the sample is known, dilute the sample with Pro-Detect™ Rapid Assay Dilution Buffer to a concentration of 1 µg/mL. If the concentration of GST protein in the sample is unknown, use a 1:25 dilution for mammalian, yeast, bacterial, and insect lysates.

For example, to make 150 µL at a 1:100 dilution, add 1.5 µL of sample (cell supernatant or lysate) to 148.5 µL of Pro-Detect™ Rapid Assay Dilution Buffer and vortex or pipette up and down to mix.

Perform test

Note: Perform all test at room temperature. To avoid condensation on the strips, allow the package to warm to room temperature for 15 minutes prior to removing the strips from the bag.

1. For each strip, add 150 µL of diluted sample to a microtiter plate or test tube.
2. Insert the lateral flow strip with arrow facing downward into the sample and wait 10-15 minutes for the color bands to appear.
3. Remove the strip(s) from the sample after 10-15 minutes of processing time. Do not exceed 20 minutes.

   A positive test will result in a red band at the control location and at the test location. Images may be acquired by photograph (camera, cell phone) or imaging equipment. Lateral flow assays may be further saved by placing in notebooks.

After results are obtained, if signal appears weak, perform a second dilution of 1:100 and re-test using steps 1-3 above. A weak signal can result from a high concentration of tagged protein in the protein. The additional dilution will bring the concentration into the acceptable working range. If upon detection with the second dilution no test line is observed, the original test performed was at a concentration below the recommended working range.

Troubleshooting

Observation	Possible cause	Recommended action
Low intensity at the test line.	Sample was below the recommended concentration range.	Reduce dilution (1:10) and retest using a new lateral flow strip.
	Sample was above the recommended concentration range.	Perform a second dilution on the diluted sample to bring the concentration into the recommended working range (see the Additional Information section).
No control line detected.	Lateral flow strip was not sufficiently submerged in sample.	Apply test strip fully into sample well and ensure enough volume is present in the sample well to fully cover the white application tip of the lateral flow strip. Flow of liquid up the strip should be visible within 30 seconds.
No test lines detected.	Sample did not contain protein tag.	Verify correct assay strip is being used.
		Verify presence of protein tag via an alternative method (i.e., ELISA or Western blot).
	Lateral flow strip was not sufficiently submerged in sample.	Apply test strip fully into sample well and ensure enough volume is present in the sample well to fully cover the white application tip of the lateral flow strip.

Additional information

Lateral flow assays are highly robust assays that can withstand many commonly used detergents, buffers, salts, and other lysis reagents. A list of commonly used reagents and the effective compatible concentrations are provided in the table below.

Table 1 Assay reagent compatibility.

Salts/Buffers		Detergents	
Substance	**Compatible Concentration**	**Substance**	**Compatible Concentration**
NaCl	1.5 M	SDS	0.2%
Urea	0.4 M	Triton™ X-100 Detergent	1%
RIPA Buffer	Undiluted	CHAPS Detergent	1%
B-PER™ Bacterial Protein Extraction Reagent	Undiluted	**Misc. Reagents and Solvents**	
M-PER™ Mammalian Protein Extraction Reagent	Undiluted	Glycerol	10%
KCl	1.5 M	NP40 Detergent	1%
		EDTA	5 mM

Manufacturer: Thermo Fisher Scientific | 3747 N. Meridian Road | Rockford, Illinois 61101 USA