

DSMZ B-FAST ELISA

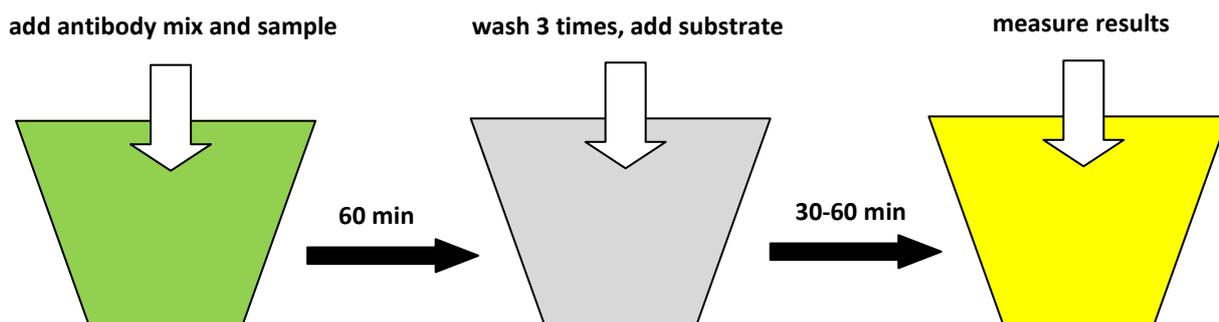
Detection of plant viruses

This kit (96 tests) contains a frame with 12 8-well strips, 2 vials with each 1.4 ml capture antibody, 2 vials with each 1.4 ml detection antibody and this protocol.

Protocol:

- 1: **Mix equal amounts of capture and detection antibody, add 50 µl of this antibody mix to each well.** (e.g. mix 0.2 ml of capture and 0.2 ml of detection antibody for one strip of 8 wells)
 - 2: **Add 50 µl aliquots of the test sample extracted in sample buffer (1:20 w/v) to the wells (ideally in duplicate). Incubate for 60 minutes at 37°C.** (Optional: Shaking at 100 rpm improves sensitivity!)
- (Note: If not all strips are required [less than 96 samples], please remove these strips and keep them in the sealed aluminum bag at 4°C until use! Do not forget to keep the frame for future use!)
- 3: **Wash strips three times with PBS-Tween with an ELISA plate washer (or using a wash bottle).**
 - 4: **Add 100 µl of freshly prepared substrate (10 mg p-nitrophenyl phosphate [e.g. Sigma] dissolved in 10 ml of substrate buffer) to each well. Incubate for 30-60 min at 37°C.** (Optional: Shaking at 100 rpm improves sensitivity!)
 - 5: **Assess results by spectrophotometric measurement of absorbance at 405 nm.**

Schematic protocol:



Advantages of the DSMZ *B-Fast ELISA*:

- Convenient protocol with only very few steps and hands-on time!
- No antibody dilution required!
- Compatible with standard ELISA buffers (sample buffer, PBS-Tween wash buffer, substrate) and equipment!
- Reliable results after just two hours!

Buffers required for *B-Fast ELISA*:

PBS (pH 7.4) phosphate buffered saline

8.0 g sodium chloride (NaCl)
0.2 g monobasic potassium phosphate (KH₂PO₄)
1.15 g dibasic sodium phosphate (Na₂HPO₄)
0.2 g potassium chloride (KCl)
0.2 g sodium azide (NaN₃)
Dissolve in 900 ml H₂O, adjust pH to 7.4 with NaOH or HCl and make up to 1 l.

PBS-Tween (PBST)

PBS + 0.5 ml Tween 20 per litre

Sample buffer (pH 7.4)

PBST + 2% PVP (e.g. Serva PVP-15 polyvinyl pyrrolidone)

Substrate buffer

97 ml diethanolamine
600 ml H₂O
0.2 g sodium azide (NaN₃)
Adjust to pH 9.8 with HCl and make up to 1 liter with H₂O

Important note:

The ELISA strips and antibodies of the DSMZ *B-Fast ELISA* kit are not compatible with other plates/strips or antibodies!

Questions?

Please contact us at +49(0)531 26 16-401/-402 or plantvirus@dsmz.de!