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 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 theses 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:

- add antibody mix and sample
- wash 3 times, add substrate
- measure results

60 min

30-60 min
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!