

# DSMZ B-FAST ELISA

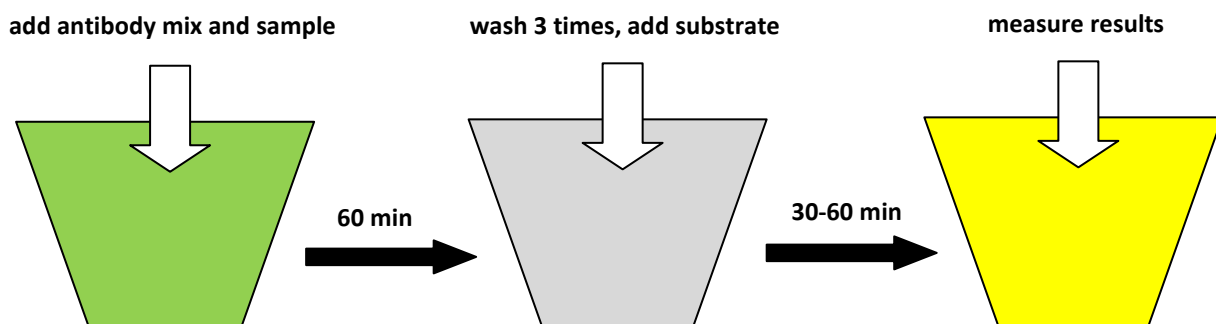
## Detection of plant viruses

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 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 ( $\text{KH}_2\text{PO}_4$ )  
1.15 g dibasic sodium phosphate ( $\text{Na}_2\text{HPO}_4$ )  
0.2 g potassium chloride (KCl)  
0.2 g sodium azide ( $\text{NaN}_3$ )  
*Dissolve in 900 ml  $\text{H}_2\text{O}$ , 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  $\text{H}_2\text{O}$   
0.2 g sodium azide ( $\text{NaN}_3$ )  
*Adjust to pH 9.8 with HCl and make up to 1 liter with  $\text{H}_2\text{O}$*

**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](mailto:plantvirus@dsmz.de)!**