XEMATest *FUSARIUM*

Rapid immunochromatographic test for qualitative determination of *Fusarium* antigen in grain and flour
Version of the test I (1902)

Fusarium genus includes highly aggressive pathogens and saprophytes which develop on weakened plants as well as endophytes coexisting with the plant with no signs of harmful effect. The diseases in various crops (cereals, legumes, beets, peas, potatoes, tomato, cucumber, rapeseed, etc.) include root rot and plant wilt caused by *F.solani, F.oxysporum, F.graminearum, F.culmorum, F.avenaceum*. Fungal infection of plant seeds causes their molding and germination loss, leading to crop thinning and yield decrease.
Fusarium fungi are able to synthesize a variety of biologically active substances, including vitamins, antibiotics and mycotoxins. The most dangerous species are: *F.graminearum* forming deoxynivalenol (DON) and zearalenone; *F.sporotrichioides, F.langsethiae, F.sibiricum* forming T-2 and HT-2 toxins*; F.verticillioides* and *F.proliferatum* forming fumonisins. Plant products containing mycotoxins may cause acute or chronic poisoning of people, animals and birds, provoking loss of appetite, dermatitis, anemia, hemorrhagic sepsis and immunosuppression. In addition, *Fusarium* fungi may also cause mycoses and allergy in humans and animals.

XEMATest *FUSARIUM* is based on polyclonal antibodies and detects multiple genus specific antigens of the fungi of *Fusarium spp*.
XEMATest *FUSARIUM* is based on immunochromatographic principle (lateral flow). The target antigen is bound by specific antibodies attached to colored microparticles. Then this complex migrates to the test line where it binds to another specific antibody to form a colored line indicating positive result.

The test is applicable for **qualitative detection** of target antigens in cereals and legumes, grain, fodder and food based on plant materials, soil samples.

TEST SENSITIVITY AND SPECIFICITY
The detection sensitivity is 20 U/ml of *Fusarium* antigen. Based on the average protein content in *F.graminearum* mycelium extracts, it approximately corresponds to 20-50 ng/ml of total protein (Lowry method) when extracting with isotonic neutral buffer (0.1M PBS, pH 7.2-7.4). This calculation is approximate and depends on the preservation of the studied material, method of storage and expression of specific antigens depending on climatic conditions, species, strain of fungus and host plant.
PLEASE NOTE: the sensitivity is calculated for the target antigenic material content in solid material extracted by the method described below, (solid/liquid ratio 1:10 wt/vol).
This test determines the antigen in the vegetative form of the fungus and practically does not determine the antigen in spores.
DETECTED low cross-reactivity (3-5%) with a species of *Trichothecium roseum*, trace cross-reactivity (0.1-1%) with species of *Aspergillus repens* and *Microdochium nivale*.
NOT DETECTED cross-reactivity (<0.1%) with the following genera and species: *Aspergillus* of other species, *Candida, Saccharomyces cerevisiae, Mucor, Rhizopus, Thamnidium, Phoma, Alternaria; Trichoderma, Phytophtora, Pythium, Penicillium, Cladosporium, Ustilago, Botrytis, Geomyces, Ascochyta, Claviceps, Neurospora, Acremonium, Ophiostoma; Trichophyton, Microspora*.
Due to extreme antigenic polymoprphism within some fungal genera it is impossible to predict the absence of cross-reactivity for given species basing on the data obtained with other species of the same genus.

SPECIES COVERAGE
The kit was tested on material obtained from pure cultures of species and species complexes listed below. The list of covered species is continuously updated, please inquire at XEMA technical support. The degree of cross-reactivity of all *Fusarium* species against reference species varies within a range 30-100%
There is no data that any species of the genus *Fusarium* is NOT determined by this test system. In case of doubtful results, it is recommended to determine the presence of *Fusarium* fungus antigen by laboratory methods (for example, XEMA ELISA, cat. N˚ K827).

FGSC – graminearum	***FFSC FOSC – oxysporum***	***FFSC – fujikuroi***
F. graminearum	*F. oxysporum*	*F.fujikuroi*
FSAMSC – sambucinum	***FSSC – solani***	*F. verticillioides*
F. sambucinum	*F. solani*	*F. proliferatum*
F. poae	***redolens species complex***	***FTSC – tricinctum***
F. sporotrichioides	*F. redolens*	*F. tricinctum*
F. cerealis	***FIESC - incarnatum/equiseti***	***FCSC – chlalmydosporum***
F. langsethiae	*F. equiseti*	*F.chlamydosporum*
FASC – avenaceum		
F. avenaceum		

More details of test performance (sensitivity, specificity, variability, influence of matrix and processing) are available on request or online at www.xematest.com

CONTENTS

- 5 test strips individually packed into sealed pouches;
- 5 Sample collection tubes;
- 2 vials of Sample extraction buffer – 26 ml each;
- 5 disposable pipettes;
- 5 Sample dilution tubes containing Sample dilution buffer – 5 ml each;
- Instruction for use.

SPECIMEN HANDLING

The specimens should be brought to temperature range +18...+35°C before use; testing of colder specimens diminishes the sensitivity of the assay; testing of hot specimens is NOT possible!

TEST PROCEDURE

1. Allow the test strips to reach room temperature for 5-10 minutes before opening the pouches.
2. Use blender or mortar to crush the material. Continue crushing until a homogeneous powder is obtained. **Attention! If different products are analyzed, wash your crushing device thoroughly to avoid cross-contamination!**
3. Put a small piece of tested material (0.1-0.5 g) into a Sample collection tube. If the balances are available, record the actual weight in grams.
4. If the accurate volume measurement tool (pipette) is available, calculate required Sample extraction buffer volume as 10x actual weight (for example, if the weight is 0.1 g, add 1 ml of the buffer). Otherwise, add not more than half of tube volume of Sample extraction buffer. Adjust the sensitivity of the assay if the weight/volume ratio is different from 1:10.
5. Screw the cap securely onto the tube. Shake it for 5 minutes.
6. Centrifuge the extracts 5 min at 200 g to remove particles. If the centrifuge is not available, clear the solution by filtering through a tissue patch or let set down for 10-15 minutes. The clarified extract can be used immediately; not recommended to store extracts in liquid form for more than 1 hour. For longer storage place the extract to a freezer (below -15 °C).
7. Using the disposable pipette provided add TWO droplets (or 50 ul) of the extract into the Sample dilution tube and mix the content thoroughly.
8. Open the pouch carefully, taking care not to damage the test strip. Dip the test strip into the diluted extract without touching the precipitate at the bottom.
9. Allow the strip to remain in the solution for 5 – 10 seconds. Remove the test strip and place onto a clean horizontal surface; do not touch or move the test strip for 10 minutes. Read the test result.

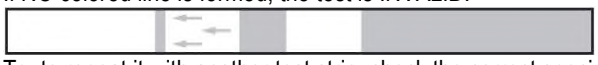
CAUTION: ENSURE THE TEST STRIP IS DIPPED AS SHOWN IN A FIGURE. THE DIRECTION AND THE DEPTH OF IMMERSION MUST BE AS SHOWN FOR CORRECT OPERATION.

INTERPRETATION OF THE RESULTS

Test is considered POSITIVE if TWO colored lines appear in the test zone.

Test is considered NEGATIVE if only ONE colored line is clearly visible.

If NO colored line is formed, the test is INVALID.



Try to repeat it with another test strip, check the correct specimen handling and test procedure, expiry date and storage conditions.

PRECAUTIONS

- The test strips should be stored at +10...+30 °C.
- Use the test within 10 minutes after opening the pouch because the test strips are very sensitive to moisture.
- Do not touch the reaction membrane.
- Do not use the test strip if its pouch is torn, or test strip is broken or damaged.
- The test strips are disposable; do not use them repeatedly.
- Do not use the test strips beyond the expiration date.

MANUFACTURER:
Xema group of companies
www.xematest.com