SYMMETRIC M1 BF

Lateral Flow Test Kit

for the quantitative detection of Aflatoxin M1 in cow, sheep, goat and UHT milk. In accordance with the EU legislation limit for baby food.

This Lateral Flow test kit is manufactured by ProGnosis Biotech S.A.

ProGnosis Biotech S.A. is ISO 9001:2015 certified by TÜV Hellas (TÜV NORD).

Use only the current version of Product Data Sheet enclosed with the kit.

Symmetric M1 BF, S2648/S2696, is a Lateral Flow Test kit for the quantitative determination of Aflatoxin M1 in cow, sheep, goat, UHT milk.

This kit contains all reagents required for 48 or 96 reactions.

- Test Time : 10min
- Range: 5 - 100ppt
- Limit of detection (LOD): 3.8ppt
- Limit of quantification (LOQ): 5ppt
- Shelf life: 12 months

This is an electronic version, please verify always the last one included in the kit.

1. Description

Symmetric M1 BF is an innovative Lateral Flow test, utilizing state-of-the-art features for the quantitative detection of Aflatoxin M1 in cow, sheep, goat and UHT milk.

2. General Information

Aflatoxins are toxic metabolites of major concern to the dairy industry, generally produced by Aspergillus flavus, A. parasiticus and A. nomius. They can have immunosuppressive, mutagenic, teratogenic and carcinogenic effects. Aflatoxins that are ingested by animals in contaminated pellets and forage are bio-transformed at the hepatic level into Aflatoxin M1 (AFM1). Aflatoxin is then excreted in this form into the milk used for human consumption and, it is also present in dairy products. AFM1 in milk and milk products is considered to pose certain hygienic risks for human health and as a result there is an established EU limit of 0.05 μg/kg (50 ppt) and the limit for baby food is 0.025 μg/kg (25 ppt).

3. Principle of the Method

The quantitative lateral flow test is based on the immunochromatography assay principles. The wells of the microtiter strips contain AFM1 specific antibodies conjugated to colloidal gold. Cow, sheep or goat milk samples are added into the wells and the suspended mixture is incubated. During this step, AFM1 (if it is present) binds to the antibodies. A test stick with two lines, capture (test) and control, is dipped into the mixture. The liquid starts flowing vertically on the test stick and passes through the two lines. A valid test should always have the upper control line red. If the sample is free of AFM1, a color development occurs at the test line, indicating the absence of AFM1 in the milk sample. On the contrary, the presence of AFM1 in the sample will cause a reduced colored signal at the test line. The intensity of the test line color is inversely proportional to the concentration of AFM1 in the samples. By utilizing S-Flow software AFM1 is accurately quantified.

4. Reagents Provided

Symmetric M1 BF kit contains sufficient reagents and materials for 48/96 measurements.

- 6/12 containers each with 1 strip of 8 reagent microwells and 8 test sticks.

5. Materials required but not provided

- 100 or 200 μl adjustable single channel micropipettes with disposable tips
- **S-Flow** software along with matching scanner device provided by lateral logic ltd
- **One-touch** Incubator for strips with automatic release

6. Storage Instructions

Store kit components between 2 - 8°C (36 - 46°F). Do not freeze any components provided. Reseal the unused strips in the storing tube together with the desiccant bag provided. The expiry date of the kit and reagents is stated on their labels and no quality guarantee is accepted after the expiration date. The expiry of the kit components can only be guaranteed if the components are stored properly and the reagent is not contaminated due to prior handling. Do not interchange individual components between kits of different lot numbers.

7. Safety and Precautions for use

All reagents should be brought to room temperature (18 - 24°C) before use (at least half an hour) and covered when not in use. Use a clean disposable plastic pipette tip for each reagent, to avoid cross contamination.

8. Method Procedure

1. Place the equipment of automatic release (test top) on the incubator and connect it to the appropriate input, located at the left side of the incubator. Plug in the **One-touch** Incubator and wait until the temperature has been stabilized at 40°C.
2. Before opening the reagents, take the kit out of the fridge (at least for half an hour) and wait until the temperature of the reagents reaches the ambient temperature.
3. Open one plastic container and take out as many test sticks and microwells as milk samples to be tested (tests per experiment ≤ reader positions). If needed, using scissors, carefully cut the number of reaction wells.
4. The container with test sticks should always be well closed after reagents have been taken out. - A container with test sticks should be emptied before another is opened.
5. Shake the milk samples vigorously or vortex.
6. Place the microwell(s) in the incubator
7. Place a new tip on the micropipette and dispense **100μl** of milk into each of the microwells. Using the same pipet tip, aspirate the sample up and down about 10 times to completely mix the lyophilized gold particles in the milk, while avoiding bubbles. The sample should turn into a **uniform pink color**. After mixing the particles, remove and discard the pipet tip. In case of more than 3 samples, an 8 channel multipipette should be used.

   The ideal temperature of the milk sample is between 4 and 18° C.
8. Push the START(RUN) button. The 5-minute countdown starts.
9. Place the appropriate number of test sticks into the automatic release equipment.
10. When the 5 minutes are over, the stick will be automatically dropped into the microwells and the **second incubation** of 5 minutes will start. **Always check if the test sticks have been dropped into the wells.**
11. When the 5 minutes of the second incubation are over, i.e. after the sound-signal, press START (STOP)* again to stop the ringing tone and take the test sticks out of the microwells.
12. Remove the white cotton sample-pad of the stick. Hold the stick from the top and remove the white sample-pad with your hands. Do not use a paper towel or any other material.
13. Place the test stick inside the plastic holder in order to be scanned. In case of S-Flow or 3PR mini reader, the sticks must be facing up.
14. Use S-flow software to quantify results as soon as possible and no later than 10 minutes after the end of analysis. Choose the type of milk analyzed. For pasteurized and fresh cow milk choose Cow. For fresh goat and fresh sheep milk choose Goat and Sheep, respectively. For Ultra High Temperature milk choose UHT. The software will use a Lot specific curve to calculate the results in parts per trillion (ppt). A simple visual interpretation of the stick is NOT possible.

9. General Specifications

- **Cross-reactivity:** The anti-Aflatoxin M1 antibody has <5% cross reactivity with Aflatoxin M2 and no cross-reactions other mycotoxins (Ochratoxin A, Zearalenone, Deoxynivalenol and Fumonisin B1) and other unrelated compounds, such as antibiotics (Benzylpenicillin, Cefalonium, Oxytetracycline, Erythromycin, Neomycin, Enrofloxacin, Sulfadiazine, Trimethoprim and Dapsone).
- **Matrices:** Raw and pasteurized cow, goat and sheep milk, UHT milk.
- **IC50** = 20 - 45ppt
- **Limit of detection:** 3.8ppt
- **Limit of quantification:** 5ppt
- Accuracy of the results (concentrations between 25 and 55ppt of AFM1) < 8% CV
- Quadruplicate value at 25ppt CV ≤ 7%

10. Interferences

There are no interferences from somatic cells at 10^6 SCC/ml or bacteria at $3x10^6$ CFU/ml.

11. Performance Evaluation

Proficiency Tests

All products participate frequently in Proficiency Tests. For more information, visit the individual product page in our website: **www.prognosis-biotech.com**

12. Summary of method

Total method time: 10 minutes

Plug in the incubator and wait until the temperature is stabilized at 40°C

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Shake vigorously the milk samples

⇩

Place the microwells in the incubator

⇩

Dispense 100μL of each sample into the microwells and mix 10 times the milk with the lyophilized gold particles

⇩

Push the START (RUN) button

⇩

Place the test sticks into the automatic release equipment

⇩

(Wait 5 mins)
The test sticks will be automatically dropped

⇩

(Wait 5 mins)
Take the test sticks out and remove the white sample-pad

⇩

Place the test sticks in the appropriate device to be scanned

⇩

Quantify through s-flow software

VERSION N2

CAT.NUMBER: S2648/S2696

STORAGE: 2-8°C

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LATERAL FLOW TEST KIT

for the quantitative detection of Aflatoxin M1 in cow, sheep, goat and UHT milk. In accordance with the EU legislation limit for baby food.

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This kit contains all reagents required for 48 or 96 reactions.

Test Time : 10min
Range: 5 - 100ppt
Limit of detection (LOD): 3.8ppt
Limit of quantification (LOQ): 5ppt
Shelf life: 12 months

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