

STORAGE: 2-8°C



for the quantitative detection of Aflatoxin M1 in cow and UHT milk

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 1000, S2448/S2496, is a Lateral Flow Test kit for the quantitative determination of Aflatoxin M1 in cow and UHT milk.

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

• Test Time: 3min

• Range: 0 - 1000 ppt

• Store: 2-8°C

• Shelf life: 12 months

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



#### 1. Description

Symmetric M1 1000 is an innovative Lateral Flow test, utilizing stateof-the-art features for the quantitative detection of Aflatoxin M1 in cow 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 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 US, India and China limit of 0.5  $\mu g/kg$  (500 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. Milk samples are added into the wells. A dipstick with two capture lines, test and control, is dipped into the mixture. The liquid starts flowing vertically on the dipstick 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 test line color intensity is indirectly proportionate to the concentration of AFM1 present in the samples. By utilizing S-Flow software AFM1 is accurately quantified.

# 4. Reagents Provided

Symmetric M1 1000 kit contains sufficient reagents and materials for 48/96 measurements.

• 6/12 pots each with 1 strip of 8 reagent microwells and 8 dipsticks.

#### 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

### 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

- Plug in the **One-touch** Incubator and wait until the temperature has been stabilized at 40°C.
- 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.
- Open one plastic pot and take out as many test strips and microwells as there are milk samples to be tested. If needed, using scissors, carefully cut the number of reaction wells.
- The pot with dipsticks should always be well closed after reagents have been taken out. - A pot with dipsticks should be emptied before another is opened.
- 5. Shake the milk samples vigorously or vortex.
- 6. Place the microwell(s) in the incubator.
- Place a new tip on the micropipette and dispense 100 μl of milk into each of the microwells.

Using the same pipette 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 $18^{\circ}$ C.

- 8. Immediately, place the appropriate number of sticks into the wells.
- 9. Push the START(RUN) button. The 3-minute countdown starts.
- 10. When the 3 minutes of incubation are over, i.e. after the sound-signal, press START (STOP)\* again to stop the ringing tone and take the dipsticks out of the microwells.
- 11. Remove the white cotton sample-pad of the stick. Touch the stick with your hand from the colorful pad and remove the white pad with your hands. Do not use a paper towel or any other material.
- 12. Place the stick inside the plastic holder in order to be scanned. In case of S-Flow scanner, the sticks must be facing up. In case of EPSON scanner, the sticks must be facing down (inverted) and the

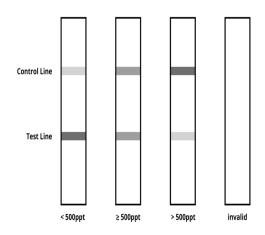
colored side must be facing the orange sticker.

13. 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 Ultra High Temperature milk choose UHT. The software will use a Lot specific curve to calculate the results (ppt). Refer to the S-Flow manual for a detailed description of the procedure. A simple visual interpretation of the stick is NOT possible.

#### 9. Visual Interpretation

When the test time is completed (3 min), the test strip can also be visually read and interpreted according to the following figure.

Visual result interpretation index



#### 10. General Specifications

- Cross-reactivity: The anti-Aflatoxin M1 antibody has no cross-reactions with analogous compounds (Aflatoxin M2), 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, UHT milk
- IC50 = 200 450ppt
- · Limit of detection (LOD): 47 ppt M1
- Limit of quantification (LOQ): 70 ppt M1
- Accuracy of the results (concentrations between 250 and 700 ppt of AFM1) < 8% CV</li>
- Quadruplicate value at 500 ppt CV (%) < 7

There are no interferences from somatic cells at 10<sup>6</sup> SCC/ml or bacteria at 3 x 10<sup>6</sup> CFU/ml.

# 12. Summary of method

Total method time: 3 minutes

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



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



Place the test strips into the wells



Push the START (RUN) button



(Wait 3 mins) Push the START (STOP) button



Take the strip out and remove the white sample-pad



Place the strip in the appropriate device to be scanned



Quantify through s-flow software

All immune assays supplied by ProGnosis Biotech S.A., are warranted to meet or exceed our published specification when used under normal conditions in your laboratory. If the product fails during the stated period, a replacement product will be issued.

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#### LATERAL FLOW TEST KIT

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e: info@prognosis-biotech.com t: +30 2410 623922 f: +30 700 700 6262 Farsalon 153 | 41335 Larissa, Greece

