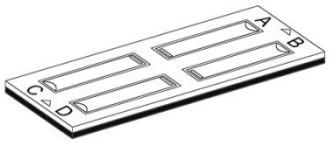
LACTOSCAN SCC KIT x4

Instruction manual

Kit consists of the following disposables:

- 100 pcs. LACTOCHIP x4 

- 400 pcs. SOFIA GREEN lyophilized dye 

- 800 pcs. tips for automatic pipette 

Preparation of sample for analysis

To prepare a sample for analysis are needed:

- Raw milk;
- LACTOSCAN SCC KIT;
- 2 pcs. automatic pipettes.
- Mini Vortex mixer

Note!

Measurement accuracy depends on the correct and consistent implementation of all stages of sample preparation and good mixing of the sample. To minimize the difference in reporting the results of several tests on the same sample of milk, always stir well before taking the sample. The sample is representative only when stirred well.

Stages:

1. **Preparation of the raw milk:**

It is mandatory a raw milk, just milked or preserved with room temperature 15-25ºC. The necessary min. volume is 30 mL.

Note!

According to **INTERNATIONAL STANDARD ISO 13366-1|IDF 148-1:2008**, to obtain the best results, you should observe the following principles:

If the samples are without preservative, they should be measured within 6 hours after milking.

If samples cannot be measured in the course of these 6 hours, they must be preserved with Bronopol ($C_3H_6BrNO_4$), Potassium Dichromate ($K_2Cr_2O_7$) or Formalin (CH_2O) in amounts specified in the standards for sampling for analysis. The final concentration of Bronopol shall not exceed 0,05 g per 100 ml of test sample. The final concentration of potassium dichromate shall not exceed 0,1 g per 100 ml of test sample. They may be stored in a refrigerator at 4 °C ± 2 °C for no longer than 6 days.

We recommend using Bronopol as a milk preservative!

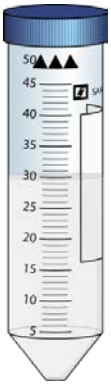
Before measuring samples, they must be heated up to 40 °C and cooled down to 20 °C and then stirred thoroughly with Vortex mixer. When samples are kept in a refrigerator, the fat globules float to the top and majority of the leukocytes adhere to them, and therefore the somatic cells go up together with fat globules. Often if the sample is not heated up to 40 °C and cooled down to 20 degrees, it is not possible to be mixed thoroughly, leading to uneven distribution of somatic cells in the sample volume. Then the measurements will vary.

Preserved and stored in a fridge sample is suitable for measuring no longer than 5-6 days.

The sample must be no more than 50 ml, and must not fill the bottle with the sample to the cap in order to allow easier mixing with Vortex mixer or by hand.

Note!
If the analysis is not made within 3-4 hours after milking, preservation of milk is required. When preserving raw milk, it is recommended to use formalin, bronopol or potassium dichromate ($K_2Cr_2O_7$).

Note!
If the preserved milk is chilled below 10 ^{0}C, it must be tempered naturally to room temperature 15-25 ^{0}C. Freshly milked milk is not necessary to be chilled or heated.



Note!
Do not use for analyses raw or preserved milk with acidity above:

- 18 °T (Therner) for cow milk
- 17 °T for buffalo milk
- 16 °T for goat milk
- 22 °T for sheep milk

Note!
In case of measurement of milk with Fat more than 5 %, for example buffalo milk, it is necessary the milk sample to be diluted with water in ratio 1:1. Then 100 µL of it is taken and added to the lyophilised dye. Adding water prevents difficulties in milk samples entry into the microfluidic chamber.

Using the Mini Vortex mixer stir the raw milk sample. For stirring, place the tip of the container in the stirrer, press and keep it pressed for 1-2 seconds, then remove it (see 1, 2, 3.). Repeat it 3-4 times paying attention during the stirring process the sample not to reach the cap of the container.


1.1


1.2


1.3

2. Pipetting 100 µL raw milk in micro-tube with SOFIA GREEN lyophilized dye:

Take one micro-tube containing SOFIA GREEN lyophilized dye, open it, and place it on the rack.

Note!
Before start working with the automatic pipettes, read carefully their Instruction Manual. Make several trials sucking and pipetting water in order to understand when exactly the first and second stop of the working button is reached. Watch the video instruction for work with automatic pipettes from the HELP menu of LACTOSCAN SCC or in the profile of LACTOSCAN on YOUTUBE: www.youtube.com/lactoscan .

Take preliminary set to 100 µL automatic pipette. Make sure that the front cone of the pipette is clean. Place it vertically over one of the tips on the working rack and place the cone of the pipette in the opening of the tip by slightly pressing it.

From initial position (see 2.1), press the working button of the pipette till the first stop is reached (see 2.2), keep it pressed and dip 2-3 mm of the tip in the milk (see 2.3). Smoothly release the working button and take out the tip from the liquid. Touch the walls of the bottle to remove the excessive milk (see 2.4, 2.5).


2.1


2.2


2.3


2.4


2.5

Pipette milk in the opened micro-tube on the rack by smoothly pressing the working button of the pipette from the initial position to the first stop (see 2.6, 2.7). After a short period press the button to the second stop (see 2.8). In this way you'll empty the tip and will guarantee precise pipetting. Always pipette the milk without dipping the tip in the lyophilized dye.

Release the button to its initial position (see 2.9).


2.6


2.7


2.8


2.9

3. Stirring the sample:

Close the micro-tube containing SOFIA GREEN dye and milk sample. Take it from the rack and place the tip of the micro-tube in the opening of the stirrer Mini Vortex. Press and hold it pressed for 1-2 seconds and remove. Repeat 8-9 times being careful while stirring the solution not to reach the cap of the micro-tube. (see 3.1, 3.2 and 3.3)


3.1


3.2


3.3

4. Interaction of milk with dye:

1 minute is needed for this interaction. If it is less 1 minute or more than 20, the analysis result may be with deviation 2-3%.

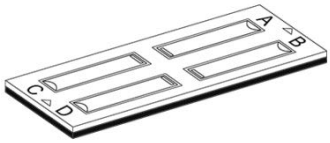
5. Repeated stirring the sample:

Take the micro-tube containing the sample from the rack and place its tip in the opening of the stirrer Mini Vortex. Press and keep it pressed for 1-2 seconds, remove. Repeat 2-3 times, paying attention place the tip of the container in the stirrer, press and keep it pressed for 1-2 seconds, then remove (see 3.1, 3.2, 3.3.). Repeat it 3-4 times paying attention during the stirring process the sample not to reach the cap of the container.

Note!
If more than 5 minutes are passed after the milk was placed into the micro-tube containing SOFIA GREEN dye, before filling the LACTOCHIP, the sample must be re-stirred using the Mini Vortex mixer.

6. Pipetting 8 µL sample in the microfluidic camera of the LACTOCHIP x4:

Open one LACTOCHIP x4.



Note!
Do not touch the upper surface of the LACTOCHIP x4. Always hold its side edges.

Take preliminary set to 8 µL automatic pipette. Make sure that the front cone of the pipette is clean. Place it over one of the tips on the rack and place the cone of the pipette in the opening of the tip by slightly pressing it.

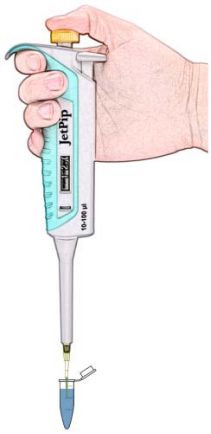
Open the micro-tube containing the sample.

From initial position (see 6.1), press the working button of the pipette till the first stop is reached (see 6.2), keep it pressed and dip 2-3 mm of the tip in the sample (see 6.3). Smoothly release the working button to the initial position. Take out the tip from the liquid. Touch the walls of the bottle to remove the excessive milk (see 6.4, 6.5).

6.1

6.2

6.3



6.4

6.5

Now there's 8 µL sample in the tip.

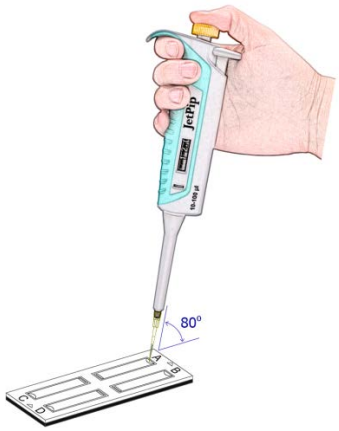
Take the LACTOCHIP x4 by holding its side edges.

Pipette the sample at an angle of approximately 80º to the filling opening in semicircular shape. Pipetting is done by smooth pressing the working button of the pipette from the initial position to the first stop (see 6.6, 6.7) Hold the button at the first stop, remove the pipette from the LACTOCHIP and smoothly release the button to the initial position (see 6.8).

In this way you'll empty the tip and precise pipetting will be assured. Release the button to its initial position.

Note!
Do not use the second stop in order not to enter air in the micro-fluidic camera.

Note!
Avoid forming bubbles in the micro-fluidic camera and splashes during pipetting the sample.



6.6

6.7

6.8

In order to load the rest of the micro-fluidic cameras of the LACTOCHIP x4, repeat the procedure described in points 1 to 6 by consecutively filling the micro-fluidic cameras from A-D.

Note!
It is recommended to use the 4 micro-fluidic cameras at once. If you use only 1 or 2, store the LACTOCHIP x4, paying attention not to contaminate it with dust or other pollution as it will lead to false results of the analyses.

Note!
The loaded LACTOCHIP x4 must be left for 30 – 60 seconds before placing it in the device. This time is required for stopping the cells' movement inside the micro-fluidic cameras.

Note!
It is recommended to place the loaded LACTOCHIP x4 in the device and to start analyses within 1 minute. Delay may lead to inaccurate results due to evaporation of the sample and air entering it.

7. Starting analysis:

Place the loaded with sample LACTOCHIP x4 in the cartridge of the LACTOSCAN SCC. Using the software, start the analysis.

8. Disposal:

Using the button for removing the tip, leave the tip inside the micro-tube with the sample. Dispose the micro-tube with the sample residue, the tip and used for analysis LACTOCHIP x4 in suitable container.

Storage:

Store LACTOSCAN SCC KIT x4 at a temperature of -10℃ to +40℃ and shortly to -20℃, protected from direct sunlight.

For more information and video instructions how to work with LACTOSCAN SCC KIT x4 see www.lactoscan.com or www.youtube.com/lactoscan

Shelf life: see the label on the box

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