

# ECLIPSE 100

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**Test para la detección de sustancias  
antibacterianas en leche**

*Test for detection of inhibitory substances in milk*

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## SCOPE

ECLIPSE is a test for detection of inhibitory substances in milk. This is a rapid and simple method to check whether milk contains antibiotics at concentrations exceeding the Maximum Residue Limits (MRL).

## PRINCIPLE

**ECLIPSE** is based on the inhibition of microbial growth. The kit is presented in a microplate format, where each well contains agar medium spread with *Geobacillus stearothermophilus* spores plus a pH indicator.

When the plate is incubated at 65°C, spores germinate and cells grow producing acid and changing the agar pH. Changes of pH will produce changes of the agar colour, from blue (purple) to green-yellowish.

When milk samples contain antibiotic at higher concentrations than the detection limit, microorganisms will not grow neither colour changes will be observed.

## KIT COMPONENTS

- 96 individual tests microtiter plate.
- Adhesive sheet to seal the plate.
- Product certificate

## STABILITY AND STORAGE

The kit components should be stored at 4 - 12 °C and protected from light. This kit is stable for 9 months when stored under optimal conditions. See the kit shelf-life on the package.

## ADDITIONAL MATERIAL (NOT PROVIDED)

- Micropipettes
- Block heater (FX Incubator, ref: ZE/FX), water bath, or oven at 65°C
- Milk free of antibiotics to be used as negative control
- Plastic frame as holder for the wells (if necessary)
- Positive control - freeze dried Penicillin G (optional)

## SAFETY

- Protective clothing should be worn and skin contact with the reagents avoided.
- Do not ingest.
- Good laboratory practices should be used.
- A SAFETY DATA SHEET is available from your local distributor by request.

## NOTES

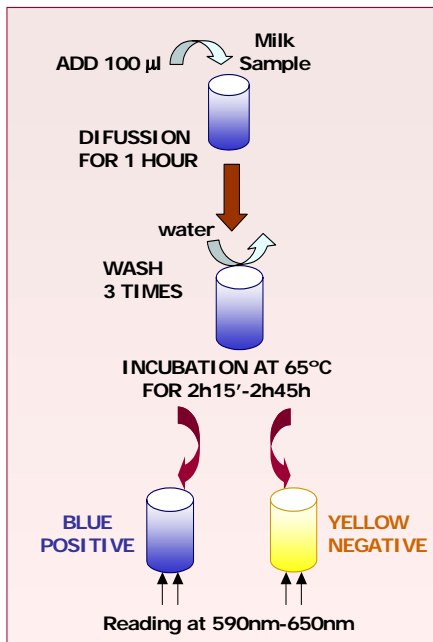
- It is strongly recommended to use a **negative control sample** (antibiotic-free milk) in each assay, to determine the optimal incubation time of the assay. It is also advisable to include a **positive control sample** (milk sample with high concentration of antibiotic (ie. penicillin G 10 mg/L). Samples used as negative or positive control must belong to the milk specie tested. A new pipette tip should be used for each sample.
- This test is extremely sensitive to antibiotics and other antibacterial substances such as detergents and disinfectants. Any contamination with these substances should be prevented.
- Although natural inhibitors contained in milk do not interfere with the test results, samples from colostrum, milk from the end of the breeding period, and mastitis milk have high concentration of these inhibitors and can alter the final results.
- When samples containing preservatives (i.e. Azidol) are analysed, bear in mind that the incubation time could increased few minutes. Please, contact ZEU-INMUNOTEC for a specific assay procedure when testing samples containing azidol.

**TEST PROCEDURE**

1. Cut the foil sheet covering the wells and split the strips to be used by pressing up from the bottom of the wells.

The foil covering the remaining wells should not be removed and the remaining wells kept immediately in a closed plastic bag at 4-12°C to prevent the agar from drying up. Although single wells can be used, it is recommended to use strips of 8 wells each time.

2. Peel off the adhesive foil covering the plate, and add **100 µl** of milk sample to each well. It is recommended to use automatic micropipettes.
3. Leave the wells/plate at room temperature (approximately 20-24°C) for **1 hour** to allow the sample to diffuse through the well.
5. **Wash** the plate/wells with distilled water by adding 300 µl of distilled water, when using a micropipette, or filling the well up, when using a squeeze bottle. Repeat the washing step 3 to 4 times. Empty the wells/plate by turning the plate upside down on top of an absorbent paper to remove the water in excess.
6. Seal the wells/plate carefully with an adhesive sheet and **incubate** at **65°C** until the negative control sample has turned yellow (approximately 2h15min - 2h30min). See the product certificate and use the incubation time shown as reference.


**7. Results:**

-Visual reading of results: turn the plate upside down and identify the **blue-violet** wells as **positive** and **yellow** wells as negative.

-Photometric reading: Read the wells/plate at **590 nm** (Filter 1) and at **650 nm** (Filter 2). Results will be interpreted as the difference between the values of the two readings. Samples with results  $\geq$  to those obtained for the samples used as negative control plus 0.3 will be positive.

POSITIVE:  $AM\ 590nm - AM\ 650nm \geq AN\ 590nm - AN\ 650nm + 0,3$

AM: Sample Absorbance

AN: Negative Control Absorbance

**NOTE: The above procedure is only applicable when the difference of absorbance at 590nm - 650nm of the negative control sample is less than 0.5.**

**Detection limits ( $\mu\text{g/ml}$ ) of ECLIPSE 100 kit for different inhibitors in cow milk:**

ECLIPSE 100	NEGATIVE	POSITIVE
PENICILLIN G	0.002	0.003
AMPICILLIN	0.003	0.004
AMOXICILLIN	0.003	0.004
OXACILLIN	0.010	0.015
CEPHALEXIN	0.050	0.075
CEPHAPIRIN	0.006	0.008
OXYTETRACYCLIN	0.050	0.100
TETRACYCLINE	0.050	0.100
SULFATHIAZOLE	0.025	0.075
SULFAMETAZINE	<0.100	0.150
SULFANILAMIDE	<0.300	0.500
ERYTHROMYCIN	0.100	0.300
TYLOSIN	0.050	0.080
NEOMYCIN	0.100	0.500
GENTAMICIN	0.100	0.300
STREPTOMYCIN	<2.000	3.000
LINCOMYCIN	0.100	0.300

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**ECLIPSE** is an *in vitro* diagnostic kit for the screening of milk samples. In analysis implicating legal processes, the results should be reevaluated with an official reference method. ZEU-INMUNOTEC, does not assume any legal responsibility

Please, do not hesitate to contact Z.E.U.-INMUNOTEC or your local distributor if any anomaly is observed .

### **OTROS TESTS PARA LA INDUSTRIA LECHERA**

- Kit RC para la detección y cuantificación de leche de vaca y cabra en leche o queso de oveja mediante la técnica ELISA.
- Kit IC para la detección rápida de leche de vaca y de cabra en leche o queso de oveja. ¡5 minutos!
- CALOKIT para la detección de calostro en leche y cuantificación de proteína sérica.
- PROTEÓN para la detección de proteínas vegetales en leche y productos lácteos mediante la técnica de ELISA.

### **OTHER TESTS FOR THE DAIRY INDUSTRY**

- RC test: ELISA test for the detection and quantification of cow or goat's milk in sheep's milk or cheese.
- IC test: Immunochromatographic test for a rapid detection of cow or goat's milk in sheep's milk or cheese). Only 5 minutes!
- CALOKIT: ELISA test for colostrum detection in milk and quantification of whey proteins.
- PROTEÓN: ELISA test for detection of vegetable proteins in milk and dairy products.

## **Web Site:**

[www.zeu-inmunotec.com](http://www.zeu-inmunotec.com)