

TYLOSIN ELISA KIT

Tylosin is macrolide antibiotic widely used in animal breedings, available both for oral and injectable use. In case withdrawal time is not respected, meat and milk can be contaminated by tylosin residues. High residue levels can also be due to poor clearance of the drug occurring in case of liver for kidney disfunction or to excessive dosages. Tylosin can also be used as a growth promoter; EU legislation banned this practice since 1999 (Council Regulation n. 2821/98), but in the USA and many other countries tylosin is often used as feed additive.

According to the EU legislation, the use of tylosin and any other antibiotics in honey bees is not permitted. During 2004 and 2005 contamination of honey with tylosin residues has been reported worldwide. In the USA the maximum residue limit for animal tissues is 200 ppb, for milk is 50 ppb. In the EU the limit for milk is also 50 ppb, while MRL for animal tissues (all food producing species) is 100 ppb (Council Regulation n. 2377/90). For honey, the Italian Ministry of Health has established an action level of 5 ppb for tylosin residues.

Assay principle

Competitive enzyme immunoassay.

Applications

Tissues (muscle, liver, kidney), raw milk, honey and urine.

Standard range curve

0,5 – 30 ng/ml.

Detection limits

- Tissues, raw milk: 5 ppb.
- Honey: 1 ppb.
- Urine: 12,5 ppb.

Measuring range

- Tissues, raw milk: 5 – 300 ppb.
- Honey: 1 – 60 ppb
- Urine: 12,5 – 750 ppb

Specificity (% di cross-reattività)

Tylosin A	100
Tylosin D	94
Spiramycin	10
Leucomycin	0.38
Oleandomycin	< 0.01
Tilmicosin	< 0.01
Erythromycin	< 0.01

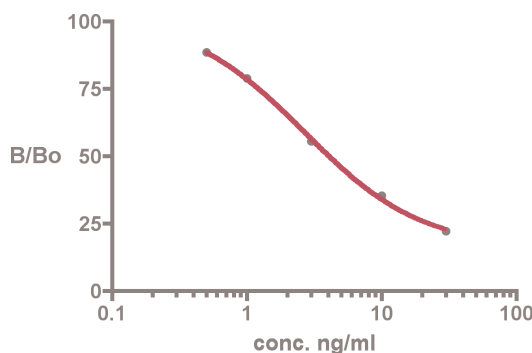
Sample preparation

- Tissues: homogenization, water extraction, centrifugation.
- Raw milk: centrifugation, dilution.
- Honey: extraction with acetonitrile, centrifugation, evaporation.
- Urine: dilution.

Assay time

50 min. (sample preparation not included).

Example of calibration curve



Advantages

- Measuring range fitting with regulatory limits.
- Easy sample preparation.
- High sensitivity.
- Applicable to many different matrices.
- Available in 96 and 48 wells format.
- Free spreadsheet downloadable at www.tecnalab.com to calculate sample concentration.

Storage conditions

+2/+8 °C.

Shelf-life

8 months from date of manufacturing.

(rev. 23/11/06)

Kit contents

- Microtiter 96/48 well plate, 12/6 strips with 8 breakable wells.
- Tylosin standard solution, concentrated.
- Enzyme conjugate , concentrated.
- Enzyme conjugate diluent, ready to use.
- Dilution buffer, concentrated.
- Washing buffer, concentrated.
- Developing solution, ready to use.
- Stop solution, ready to use.
- Instructions.

Manufacturer

Tecna Srl, ISO9001/UNI EN ISO 9001 – Ed. 2000 (SGS, N°. IT01/0291).

Materials not provided

- Distilled water.
- Ice.
- Acetonitrile.
- Sodium chloride.

Additional equipment

- Meat grinder (tissues).
- Balance (tissues).
- Centrifuge possibly refrigerated (force required: 3000 xg for raw milk; 2000 xg for tissues).
- Filter paper Whatmann nr. 4 or equivalent (tissues).
- Homogenizer or rotating wheel (tissues).
- Evaporation apparatus (honey).
- Warm bath (optional for honey).
- Micropipette 20–200 µl and 200–1000 µl with suitable tips.
- Multichannel micropipette 20–300 µl with suitable tips and reagent boats (optional).
- ELISA plate or strip reader with a 450 nm filter.