Determination of D- and L-Lactic acid in food products (without differentiation)
Test-kit for 32 determinations on the RIDA®CUBE SCAN instrument (340 nm)

For in vitro use only
Store between +2 and +8°C

Principle

Enzymatic test with D- and L-Lactate Dehydrogenase (D-LDH and L-LDH). NADH is produced and is measured at 340 nm:

L-Lactate + NAD^+ —— L-LDH ⟶ Pyruvate + NADH +H^+

D-Lactate + NAD^+ —— D-LDH ⟶ Pyruvate + NADH +H^+

Reagents

1: 32 tubes with approx. 800 µl reagent 1 (buffer)

2: 32 caps with approx. 200 µl reagent 2 (enzyme)

3: one RFID card (Radio Frequency Identification)

The reagents are stable up to the end of the indicated month of expiry, if stored at 2 - 8 °C. Do not freeze the reagents. Let the reagents reach the laboratory temperature before use (20 - 25 °C).

The general safety rules for working in chemical laboratories should be applied. Do not swallow! Avoid contact with skin and mucous membranes.

This kit may contain hazardous substances. For hazard notes on the contained substances, please refer to the appropriate material safety data sheets (MSDS) for this product, available online at www.r-biopharm.com. After use, the reagents can be disposed of with the laboratory waste. Packaging materials may be recycled.

Sample preparation

- Use clear, colourless and pH-neutral liquid samples directly, or after dilution into the relevant measuring range (see test performance)
- Filter or centrifuge turbid solutions
- Degas samples containing carbon dioxide
- Clarify samples containing proteins or fat with Carrez clarification
- Crush and homogenize solid or semi-solid samples and extract with water; filtrate or centrifuge, or use Carrez clarification if necessary
- For fat containing samples, weigh sample into a volumetric flask (min. 50 ml) and extract with hot water; cool to allow the fat to separate (for example on an ice bath for 15 min); make up to the mark with water, remove the fatty layer on the top and filter the aqueous part before testing
- Adjust the pH to approx. 8.0 by adding KOH / NaOH to acidic samples or by adding HCl to alkaline samples
- Strongly coloured/polyphenol containing samples can lead to a creep reaction and should be pre-treated before testing.

Assay specifications

The assay specifications are saved on the RFID card and are executed automatically by the instrument.

Wavelength:	340 nm
Temperature:	37 °C
Calibration:	calibration curve is saved on RFID card
Test sequence:	sample + R1 / mix / 2 min / A1 / R2 / mix / 10 min / A2
Sample volume:	20 µl (basic) or 100 µl (sensitive) The required volume should be pipetted precisely into reagent 1 (test-tube).

Handling procedure

Place the RFID Card on the instrument	
Enter sample data into tablet app.: - identification - volume (20 µl or 100 µl)	
Pipette the sample into the test-tube (reagent 1)	 
Close the tube with the cap (reagent 2), insert it into the instrument and close the door	 

Test performance

Measuring range

The results are given in mg/l by the instrument, and following ranges are recommended:
- from 35 to 1250 mg/l for the basic application (20 µl)
- from 10 to 250 mg/l for the sensitive application (100 µl)

The sample volume is 20 µl or 100 µl. For the sensitive application, it is also possible to pipette any dilution with 100 µl total volume (for example 50 µl sample and 50 µl water). Results must be recalculated accordingly.

Specificity

The test is specific for D- and L-Lactic acid. Interferences were identified for Ascorbic acid, Hydroxybutyric acid and Sulfite (SO_2) starting from 0.02 g/l. Oxalic acid interfered above 0.2 g/l, and all other measured substances did not interfere up to 20 g/l.

Notes

1. Use a quality control every day where a run is performed (e.g. Enzytec Multi-acid Standard E1240). If the deviation of this quality control is higher than 10%, it is necessary to measure the reagent blank with a water sample, and to subtract it from all future samples results.

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