

# Manual Procedure

Automated procedure on request

**MEDICHEM**  
MIDDLE EAST  
Clinical Chemistry Reagents  
Liquid Stable Reagents

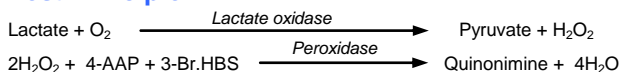
**Cat. No. 12500** R 1 x 10 ml  
For 10 tests

## Lactate

Fully enzymatic method

### Liquid Reagent

#### Test Principle



Lactate oxidase catalyzes the oxidation of lactic acid to pyruvate and hydrogen peroxide. Peroxidase then catalyzes the reaction of hydrogen peroxide with a hydrogen donor, in the presence of 4-Aminoantipyrine, to form a dye. Color intensity, measured at 510 nm, is proportional to the lactate concentration in the sample.

#### Concentrations in the test

|   |       |        |
|---|-------|--------|
| <b>Reagent R</b>                                      |       |        |
| TRIS buffer pH = 7.5                                  | 100   | mmol/L |
| 4-AAP (4-Aminoantipyrine)                             | 0.3   | mmol/L |
| Peroxidase  | ≥ 1   | KU/L   |
| Lactate oxidase                                       | ≥ 0.7 | KU/L   |
| 3-Br.HBS  | 0.27  | mmol/L |
| Surfactant, stabilizer and preservative               |       |        |
| <b>Standard</b> : Concentration as indicated on vial. |       |        |

#### Stability of reagent

**Reagent R:** liquid, ready to use.

The reagent is stable up to expiry date given on the label when stored at +2 → +8 °C. Stability after opening the bottle: 3 months at +2 → +8 °C.

#### Specimen collection and handling

1. Plasma collected on fluoride oxalate, fluoride EDTA, or sodium fluoride is the recommended specimen.
2. Blood sample should be drawn from a stasis-free vein.
3. Cells in blood samples contribute to glycolysis which increases lactate levels rapidly so, their quick removal is essential for accurate lactate analysis
4. Heparinized or EDTA plasma could be used, but precautions must be taken to retard glycolysis by keeping the whole blood on ice and then separating the plasma from the cells within 15 minutes of collection.
5. Don't use serum.
6. Cerebrospinal fluid (CSF) may be used as obtained.
7. Separated plasma is stable for 2 hours at 20 - 25°C, 2 days at 2 - 8°C, or 1 month at - 20 °C.
8. CSF is stable for 3 hours at 20 - 25 °C, 24 hours at 2 - 8°C, or 1 month at - 20 °C.

#### Calibrator / Standard

MediCal U Cat. No. 15011  
Lactate STD. Cat. No. 16141

#### Quality control

Meditrol N Cat. No 15171  
Meditrol P Cat. No 15181

#### Procedure

|                   |                           |
|-------------------|---------------------------|
| Wavelength        | Hg 546 nm ( 495 - 550 nm) |
| Spectrophotometer | 510 nm                    |
| Cuvette           | 1 cm light path           |
| Temperature       | 37°C                      |
| Measurement       | against reagent blank     |
| Reaction          | end point                 |

#### Assay

|  | Blank   | Calibrator / Standard | Sample  |
|--|---------|-----------------------|---------|
| Distilled water  | 10 µl   | --                    | --      |
| Calibrator / Standard  | --      | 10 µl                 | --      |
| Sample   | --      | --                    | 10 µl   |
| Reagent R  | 1000 µl | 1000 µl               | 1000 µl |
| Mix, incubate for 5 min. at 37 °C, or 10 min. at 25°C, Read the absorbance (A). The final color is stable for at least 30 min. |         |                       |         |

#### Calculation

$$\text{Conc. Lactate (mg/dl)} = \frac{A_{\text{Sample}}}{A_{\text{Cal./STD}}} \times \text{Conc. Cal./STD (mg/dl)}$$

#### Linearity

Up to 75 mg/dl.

Sample with values greater than 75 mg/dl, must be diluted (1+1) with saline and re-assayed. Multiply the result by 2.  
mmol/L X 9.01 = mg/dl

#### Interferences

For a comprehensive review of drug interference on lactate levels see Young et al.

#### Precautions

1. Reagents contain sodium azide as a preservative. Upon disposal flush with large volumes of water.
2. Don't use the reagents beyond the expiry date printed on the label

#### Reference range

##### Plasma

| Age Group | Reference Range | Unit  | Notes                                    |
|-----------|-----------------|-------|--|
| New born  | < 26            | mg/dl | Plasma treated with glycolysis inhibitor |
| Adults    | < 22            | mg/dl | Plasma treated with glycolysis inhibitor |
|           | 4.5 - 19.82     | mg/dl | Venous blood                             |
|           | 4.5 - 14.41     | mg/dl | Arterial blood                           |

##### CSF

| Age Group | Reference Range | Unit  |
|-----------|-----------------|-------|
| New born  | 10 - 60         | mg/dl |
| 3 - 10 d. | 10 - 40         | mg/dl |
| > 10 d.   | 10 - 25         | mg/dl |
| Adults    | 10 - 22         | mg/dl |

#### References

1. Gutmann, I., Wahlefeld, A., Methods of Enzymatic Analysis. 2nd Ed., Academic Press, New York, 1974, 1464.
2. Tietz, N.W., Fundamentals of Clinical Chemistry, 4<sup>th</sup> Ed., W.B. Saunders Company, Philadelphia, 1996, 367.
3. Tietz, N.W., Clinical Guide to Laboratory Tests, 3<sup>rd</sup> Ed., W.B. Saunders Company, Philadelphia, 1995, 382-383.
4. Westgard, J.O., Lahmeyer, B.L., Bimbaum, M.L., Clin Chem 1972, 18 : 1334 - 1338.
5. Young, D.S., Effects of Drugs on Clinical Laboratory Tests, fifth edition 2000, AACC Press, Washington, D.C.