



L-Lactic Acid (K-LATE) Procedure for ChemWell®-T Auto-Analyser

Requirements:

- L-Lactic Acid (L-Lactate) Assay Kit (K-LATE) (provides ~ 475 assays).
- K-LATE ChemWell®-T assay file.
- Use in association with the L-Lactic Acid (L-Lactate) Assay Kit (K-LATE) product data booklet.

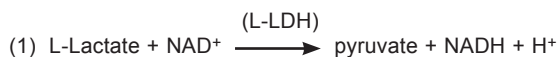
Use:

For the specific measurement of L-lactic acid especially in wines, fruit juices, beverages and food products.

For specific sample preparation methods refer to the L-Lactic Acid (L-Lactate) Assay Kit (K-LATE) data booklet.

Assay Principle:

Conversion of L-lactic acid via the following reactions is directly proportional to the coupled formation of NADH:



Procedure:

Prepare the assay reagents and calibrators and use with the K-LATE ChemWell®-T assay file.

L-Lactic Acid (L-Lactate) Assay Kit Components:

Bottle 1: Buffer (25 mL, pH 10.0) plus D-glutamate and sodium azide (0.02% w/v) as a preservative. Stable for > 2 years at 4°C.

Bottle 2: NAD⁺. Stable for > 5 years at -20°C.

Bottle 3: D-Glutamate-pyruvate transaminase suspension (1.1 mL). Stable for > 2 years at 4°C.

Bottle 4: L-Lactate dehydrogenase suspension (1.1 mL). Stable for > 2 years at 4°C.

Preparation of Kit Components:

1. Use the contents of bottle 1 as supplied. Stable for > 2 years at 4°C.
2. Dissolve the contents of bottle 2 in 5.5 mL of distilled water. **Stable for > 1 year at 4°C** or > 2 years at -20°C (to avoid repetitive freeze / thaw cycles, divide into appropriately sized aliquots and store in polypropylene tubes).
- 3 & 4. Use the contents of bottles 3 and 4 as supplied. Swirl the bottle to mix contents before use. Stable for > 2 years at 4°C.

Preparation of Assay Reagents: (per ~ 95 assays)

Reagent 1:

| Component | Volume |
|-------------------|---------|
| distilled water | 12.9 mL |
| bottle 1 (buffer) | 5 mL |
| *bottle 2 (NAD) | 1 mL |
| bottle 3 (D-GPT) | 0.2 mL |
| Total volume | 19.1 mL |

*after adding 5.5 mL of distilled water

Reagent 1 stability: > 2 days at 4°C

Reagent 2:

| Component | Volume |
|------------------|--------|
| distilled water | 2.2 mL |
| bottle 4 (L-LDH) | 0.2 mL |
| Total volume | 2.4 mL |

Reagent 2 stability: > 2 days at 4°C

Calibrators:

K-LATE 1: 0 g/L (use distilled water)
K-LATE 2: 0.25 g/L L-lactic acid
K-LATE 3: 0.5 g/L L-lactic acid
K-LATE 4: 1.0 g/L L-lactic acid

Assay Parameters:

Assay volumes: Reagent 1: 0.200 mL
Sample: 0.010 mL
Reagent 2: 0.025 mL

Calibrators: 0, 0.25, 0.5, 1.0 g/L L-lactic acid
Reaction time: 10 min at 37°C
Wavelength: 340 nm
Assay type: endpoint
Reaction direction: increase
Linearity: up to 1 g/L of L-lactic acid

