Tribioscience

L-Lactate Fluorometric Assay Kit (TBS2061, 100 Assays, Store at -20 °C)

DESCRIPTION

L-Lactate is the major stereo-isomer of lactate formed in human intermediary metabolism and is present in blood. Depending on the level of exercise, blood levels of lactate can vary between 1 and 20 mM. Lactate is generated by lactate dehydrogenase (LDH) under hypoxic or anaerobic conditions. Monitoring lactate level is a good indicator of the balance between tissue oxygen demand and utilization. L-Lactate accumulates in lactic acidosis which is associated with poor prognosis and increased mortality in patients with critical illness, sepsis and liver failure.

Tribioscience's Lactate Fluorometric Assay Kit is a simple, sensitive, and fast assay that measures the total amount of L-Lactate in biological samples. Lactate is oxidized by lactate oxidase to pyruvate and hydrogen peroxide. Horseradish peroxidase (HRP) catalyzes the reaction between hydrogen peroxide and the specific probe to yield a final product measured by the fluorometric method at Ex/Em=530/590 nm.

APPLICATIONS

Measure L-lactate in a variety of samples.

KIT CONTENTS FOR 100 TESTS:

Name	Size (100 tests)
Lactate Standard (250 µM)	100 µL
Lactate Assay Buffer	12 mL
Lactate Fluorometric Probe	40 µL
Lactate Enzyme mix	70 µL

Storage conditions: Store the Reagent at -20°C, protect it from light. Shelf life: 12 months.

PROCEDURES

1. Equilibrate all the kit components until room temperature before starting the experiment.
2. Prepare the L-Lactate standards: Add 30 µL of 250 µM L-Lactate to 270 µL of assay buffer in Tube #1, and then make a 2-fold serial dilution from Tube#2 to Tube#7) as the Table below. Tube#8 as blank control.

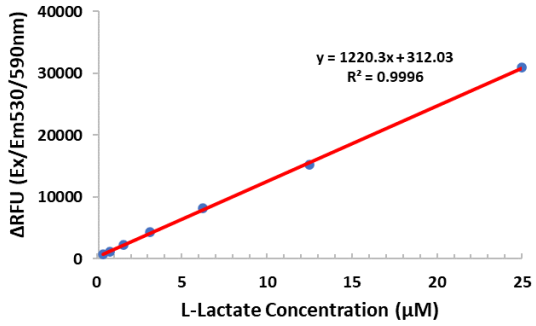
Tube #	Lactate Standard (µL)	Assay Buffer (µL)	Lactate Concentration (µM)
1	30	270	25
2	150	150	12.5
3	150	150	6.25
4	150	150	3.125
5	150	150	1.562
6	150	150	0.781
7	150	150	0.39
8	0	150	0

3. Preparation of the working solution for lactate enzyme reaction:

Component	Volume (µL) for 5 mL (1 plate)
Assay Buffer	4925 µL
Lactate Enzyme Mix	50 µL
Lactate Probe	25 µL

4. Add 50 µL of L-Lactate standards, or samples, or blank control to indicated wells in duplicate manner.
5. Add 50 µL of the lactate enzyme reaction working solution to the test samples, lactate standards and blank control.
6. Incubate at 37°C for 30 minutes with gentle shaking and protect from light.
7. Read the plate at Ex/Em = 530/590 nm for fluorometric assay.
8. The typical L-Lactate standard curve is shown in Fig. 1.

Fig. 1. L-Lactate Standard Curve



9. Calculate the sample lactate concentration by the lactate standard curve as follows:

Y=A*X + B
The lactate concentration X (µM) = (Y-B)*DF/A

Y=OD570nm; A=Slope; B=constant value; DF=dilution factor.

RELATIVE PRODUCTS

L-Lactate Colorimetric Assay (TBS2071)
LDH Cytotoxicity Assay (TBS2002)
LDH Activity Assay (TBS2012)
Resazurin Cell Viability (TBS2001)
CCK-8 Cell Viability Assay (TBS2022)
GOT Activity Assay (TBS2013)
Thiol Fluorometric Assay (TBS2026)
GSH Assay (TBS2028)
Homocysteine Fluorometric Assay (TBS2091)
AHCY Inhibitor Screening Assay (TBS2097)
G6PDH Activity Colorimetric Assay (TBS2102)
ATP Colorimetric/Fluorometric Assay (TBS2010)
ADP Colorimetric / Fluorometric Assay (TBS2020)
Caspase-3 Colorimetric Assay (TBS2030)
NNMT Inhibitor Screening Assay (TBS2097)
NNMT Activity Assay (TBS2098)

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