Intended Use
For the quantitative determination of lactate in human plasma. For in vitro diagnostic use only.

Clinical Significance
Lactate determinations are used in the diagnosis of lactate acidosis. Shock is the most widely recognized cause of lactic acidosis although, it is possible for elevated lactate levels to precede shock. Myocardial infarction, severe congestive heart failure, pulmonary edema and blood loss are the common causes of shock which will produce lactic acidosis. Lactic acidosis may also result from renal failure and leukemia. Thiamine deficiency and diabetic ketoacidosis will usually result in increased levels of lactate.

Method History
Originally lactic acid determinations were performed by either titrimetric or colorimetric methods. The first enzymatic method for lactic acid was based on the transfer of hydrogen from lactate to potassium ferrocyanide by lactate dehydrogenase (LD). This procedure was very cumbersome and did not gain wide acceptance. More current enzymatic methods involved the measurement of NADH formed from the oxidation of lactate by LD. This method has become more widely used, but still suffers from instability in many analyzer systems. The current enzymatic method is based on the action of lactate oxidase. This method is fast, accurate and is considerably more stable than previous enzymatic methods.

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-aminophenazone, to form a dye. Color intensity, measured at 550nm, is proportional to the lactate concentration in the sample.

Lactate Oxidase
Lactate + O₂ → Pyruvate + Hydrogen Peroxide

Peroxidase
Hydrogen Peroxide + TOOS + 4-AAP → Dye (550 nm)

Reagents
Lactate Reagent (R1): TRIS Buffer 100mM, 4-aminoantipyrine 1.7mM, Peroxidase (Horseradish) > 10,000 U/L, Surfactant, Stabilizer, Sodium Azide (0.09%) as preservative.
Lactate Reagent (R2): TRIS Buffer 100mM, Lactate Oxidase (Microbial) > 1,000 U/L, TOOS 1.5mM, Surfactant, Stabilizer, Sodium Azide (0.09%) as preservative.

Precautions
1. This reagent is for in vitro diagnostic use only.
2. Reagents contain sodium azide as preservative. Upon disposal flush with large volumes of water.
3. All specimens used in this test should be considered potentially infectious. Universal precautions as they apply to your facility should be used for handling and disposal of materials during and after testing.
4. Do not use the reagents beyond the expiration date printed on the kit label.

Calibration
Use an NIST-traceable lactate standard, or a suitable serum-based lactate standard. The procedure should be calibrated according to the instrument manufacturer’s instructions. If control results are found to be out of range, the procedure should be re-calibrated.
Quality Control
Reliability of test results should be routinely monitored with control materials that reasonably emulate performance of patient specimens. Quality control materials are intended for use only as monitors of accuracy and precision. The recovery of control values within the appropriate range should be the criteria used in evaluation of future assay performance. Controls should be run with every working shift in which lactate assays are performed. It is recommended that each laboratory establish their own frequency of control determination. Quality control requirements should be determined in conformance with local, state, and/or Federal regulations or accreditation requirements.

Results
To convert from S.I. units to conventional units, multiply the S.I. units by 9.01.

Example: mmol/L x 9.01 = mg/dL Lactate

Expected Values
The following reference range is suggested for L-Lactate.8

<table>
<thead>
<tr>
<th>Type</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Venous</td>
<td>0.5-2.2 mmol/L</td>
</tr>
<tr>
<td>Arterial</td>
<td>0.5-1.6 mmol/L</td>
</tr>
</tbody>
</table>

It is highly recommended that each laboratory establish its own range of expected values.

Performance
1. Assay Range: 0-15 mmol/L
2. Comparison: This lactate reagent was compared to the method performed on the Dade Chemistry Analyzer. The study was performed using 57 patient samples ranging from 0.3-10.4 mmol/L. Data was subjected to least-squares linear regression analysis which yielded a correlation coefficient(r) of 0.998 with a regression equation of y = 0.97 x + 0.1.
3. Precision: Within-Day precision for the Lactate Reagent was determined following a modification of NCCLS document EP5-T2.9 Within-Day precision studies produced the following results:

<table>
<thead>
<tr>
<th>Sample</th>
<th>N</th>
<th>Mean</th>
<th>S.D.</th>
<th>C.V.%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low</td>
<td>20</td>
<td>1.52</td>
<td>0.04</td>
<td>2.63</td>
</tr>
<tr>
<td>Mid</td>
<td>20</td>
<td>3.98</td>
<td>0.07</td>
<td>1.76</td>
</tr>
<tr>
<td>High</td>
<td>20</td>
<td>8.89</td>
<td>0.09</td>
<td>1.01</td>
</tr>
</tbody>
</table>

Day To Day precision was also determined following a modification of NCCLS document EP5-T2.9 Day to Day precision studies produced the following results:

<table>
<thead>
<tr>
<th>Sample</th>
<th>N</th>
<th>Mean</th>
<th>S.D.</th>
<th>C.V.%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low</td>
<td>20</td>
<td>1.51</td>
<td>0.04</td>
<td>2.65</td>
</tr>
<tr>
<td>Mid</td>
<td>20</td>
<td>4.12</td>
<td>0.09</td>
<td>2.18</td>
</tr>
<tr>
<td>High</td>
<td>20</td>
<td>9.19</td>
<td>0.17</td>
<td>1.85</td>
</tr>
</tbody>
</table>

4. Sensitivity: The analytical sensitivity for lactate was determined to be 0.15 absorbance units per 1 mmol/L of lactate.

References