



## ALCOHOL OXIDASE RL-100

**Description:** RL-100 is a wine-red solution of Alcohol Oxidase enzyme in buffered sucrose. The color changes reversibly to yellow in the presence of substrate under anoxic conditions. The color changes irreversibly to yellow if the enzyme is exposed to denaturing conditions.

**Identity:** Pichia Pastoris, Alcohol, Oxygen Oxidoreductase: E.C. 1.1.3.13

**Reaction:** Alcohol + O<sub>2</sub>  $\xrightarrow{\text{Alcohol Oxidase}}$  Aldehyde + H<sub>2</sub>O<sub>2</sub>

**Activity:** Alcohol Oxidase RL-100 has a guaranteed minimum specific activity of 10 EU/mg protein and 1000 EU/ml of solution as determined using the assay described in Technical Bulletin #1. One unit of activity catalyzes one micromole of ethanol to aldehyde and hydrogen peroxide per minute in an air saturated solution at pH 7.5, at 25°C.

### General

**Properties:** Alcohol Oxidase is specific for short-chain, linear aliphatic alcohols and oxidizes methanol and ethanol. The following are oxidized at a slower rate: n-propanol, n-butanol, allyl alcohol, propargyl alcohol, methyl and ethyl mercaptan, and formaldehyde. The optimum temperature range is 40 – 45°C and freezing does not inactivate the enzyme. Solubility of the enzyme is inversely related to temperature; refrigeration will often reverse cloudiness in solutions where precipitation is occurring. The pH optimum is 7.5 – 8.0 with a working range of 6.0 – 9.5. The enzyme is freely soluble in greater than 0.1 M Phosphate buffer above pH 7.0. The enzyme reversibly precipitates at below 0.05 M Phosphate and below pH 7.0. These boundaries can be extended under certain pH, temperature and ionic conditions. Alcohol Oxidase has a molecular weight of approximately 630,000 and is composed of eight subunits, each having one molecule of bound azide. Inhibitors include azide, Cu<sup>++</sup>, Ag<sup>+</sup>, Hg<sup>++</sup>, p-chloromercuribenzoate, hydroxylamine and NaF.

**Applications:** Qualitative or quantitative determination of ethanol or methanol; alcohol removal; aldehyde or hydrogen peroxide production; and oxygen scavenging.

**Formulation:** RL-100 is Alcohol Oxidase enzyme in 30% Sucrose with 0.1 M Potassium Phosphate Buffer, pH 8.0.

**Storage:** Store as a frozen solution at -20°C. Stable at sub-zero temperatures for at least one year.

**Packaging:** Alcohol Oxidase RL-100 is packaged in guaranteed minimum quantities of 20,000 and 100,000 EU. Larger quantities available upon request. Shipped with dry ice.

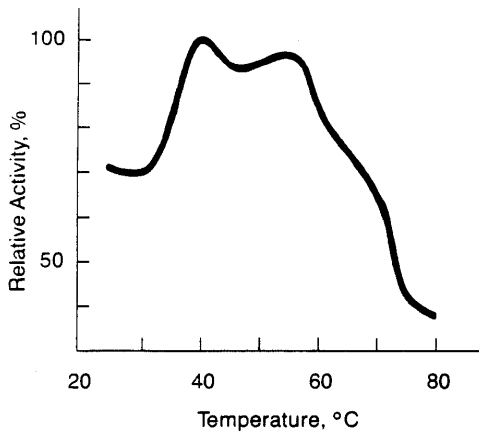


Figure 1. Temperature activity of 1:2000 dilution during 3 minutes incubation in 0.1 M phosphate buffer, pH 7.5.

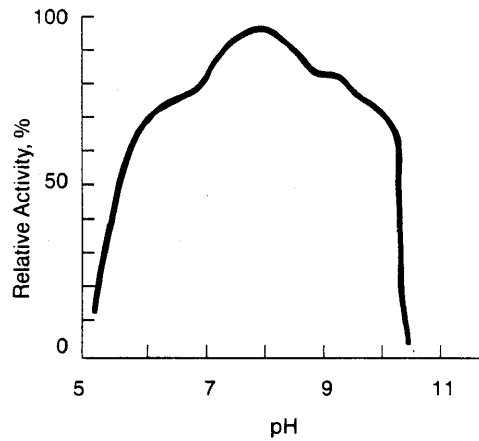


Figure 2. pH activity of 1:10,000 dilution in .025 molar citrate/ethylenediamine better determined using polarographic method at °C.

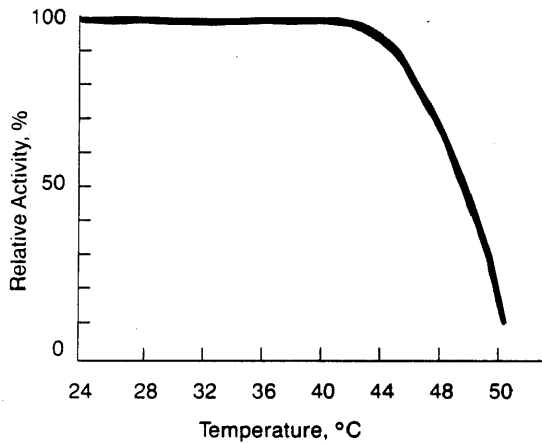


Figure 3. Thermal stability of 1:2000 dilution after 30 minutes incubation in 0.1 M phosphate buffer, pH 7.5.

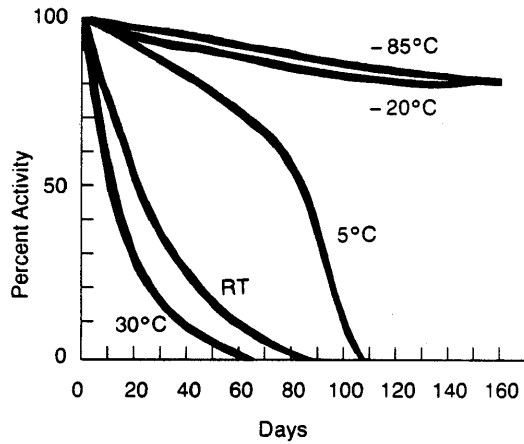


Figure 4. Long-term stability.

Relative activity (%) determined by ABTS method described in Technical Bulletin No. 1.

