

## Native Staphylococcus aureus V8 Protease, Tag Free

**Cat. No.** Protease-08S    **Lot. No.** (See product label)

### SPECIFICATION

|                               |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |
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| <b>Product Overview</b>       | Native Staphylococcus aureus V8 Protease (Endoproteinase Glu-C)                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |
| <b>Species</b>                | Staphylococcus aureus V8                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                   |
| <b>Description</b>            | Protease S. aureus V8 (Endoproteinase-Glu-C) specifically cleaves peptide bonds on the COOH-terminal side of either aspartic or glutamic acids. In the presence of ammonium, the enzyme specificity is limited to glutamic sites. It has a molecular weight of 27,000 daltons and optimum pH's of 4.0 and 7.8 with hemoglobin as the substrate. Protease S. aureus V8 is inhibited by diisopropylfluorophosphate and monovalent anions such as F <sup>-</sup> , Cl <sup>-</sup> , CH <sub>3</sub> COO <sup>-</sup> and NO <sub>3</sub> . Enzyme activity is determined by the casein digestion assay described by Drapeau. |
| <b>Tag</b>                    | Non                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                        |
| <b>Molecular Mass</b>         | 27 kDa                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |
| <b>Extinction Coefficient</b> | 4.26 (Houmard 1976).                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                       |
| <b>Optimal pH</b>             | 4.0 and 7.8 with hemoglobin substrate. (Drapeau et al. 1972).                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              |
| <b>Method</b>                 | Enzyme activity is determined by the casein digestion assay described by Drapeau (1976). Since substrate grade caseins can vary from lot to lot and according to the manner produced, the standardization of casein digestion assays has been difficult to achieve. The use of a reference enzyme preparation is suggested. One unit is that                                                                                                                                                                                                                                                                               |

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|                         | amount of enzyme which releases acid soluble fragments equivalent to 0.001 A280 per minute at 37 centigrade and pH 7.8 under the specified conditions.                                                                                                                                                                                |
| <b>Reagents</b>         | <p>1. 1% Casein in 0.05 M TrisPO<sub>4</sub> buffer, pH 7.8. (Dissolve 1 gram "Hammersten" casein in 50 ml 0.01 N NaOH with gentle heating and stirring. Add 40 ml reagent grade water and 5.0 ml 1.0 M Tris. Adjust pH to 7.8 with H<sub>3</sub>PO<sub>4</sub> and q.s. to 100 mL.)</p> <p>2. 10% Trichloroacetic acid (TCA)</p>     |
| <b>Enzyme</b>           | Dissolve at 1 mg/mL in reagent grade water.                                                                                                                                                                                                                                                                                           |
| <b>Procedure</b>        | <p>Equilibrate a series of tubes with 5.0 mL of 1% casein at 37 centigrade for 5 minutes. At zero time add 10 µL or 20 µL of enzyme. Mix. Include a reagent blank. Exactly ten minutes after adding sample, stop reaction by adding 5.0 mL TCA. Mix. Allow tubes to stand ten minutes and then filter. Read A280 of the filtrate.</p> |
| <b>Unit Definition</b>  | One Unit causes a change of 0.001 A280 nm per minute at 37 centigrade, pH 7.8 using casein as the substrate.                                                                                                                                                                                                                          |
| <b>Storage</b>          | Autolysis occurs at temperatures greater than 40 centigrade. The enzyme is fully active in USP 0.2% SDS. Stable for 12 months at 2-8 centigrade.                                                                                                                                                                                      |
| <b>GENE INFORMATION</b> |                                                                                                                                                                                                                                                                                                                                       |
| <b>Official Symbol</b>  | Protease                                                                                                                                                                                                                                                                                                                              |
| <b>Synonyms</b>         | CAS 66676-43-5; EC: 3.4.21.19; Protease                                                                                                                                                                                                                                                                                               |