1. Enzyme characteristics

1.1 Molecular form:

The metalloproteinase Aureolysin is purified from Staphylococcus aureus. Aureolysin Staphylococcus aureus is solubilized in 20 mM Tris/HCl, pH 7.8, 5 mM CaCl$_2$.

1.2 Concentration: 1 mg/ml (Pierce - BCA), 25000 units/mg using azo dye-impregnated collagen (Azocoll) as the substrate.

1.3 Purity: Aureolysin Staphylococcus aureus appears as a major protein of about 38 kDa and represents more than 95 % of total protein.

1.4 Specific activity: The protease activity was determined using azo dye-impregnated collagen (Azocoll) as substrate essentially as described (Datta, A, 1992). One unit is defined as the amount of enzyme that will release a sufficient amount of azo dye from azocoll to produce an increase in absorbance at 520 nm of 0.001 per minute at 37°C, pH 7.8.

1.5 Stability and storage: Aureolysin Staphylococcus aureus is stable if stored at -70°C or lyophilized, can be kept at -20°C for several month and at 4°C for 1 week without significant loss of activity. Repeated freezing and thawing should be avoided.

2. Applications

- Screening of inhibitors of Aureolysin Staphylococcus aureus
- Characterization of structure and function

3. Introduction to structure and function of Aureolysin

In addition to the glutamyl endopeptidase, Staphylococcus aureus secretes at least one papain-like cysteine proteinase as well as a typical metalloproteinase that is commonly referred to as aureolysin. The primary and tertiary structures of the latter enzyme have been determined (Banbula, A et al.,1998) revealing a polypeptide chain of 301 amino acids which is folded into a β-pleated N-terminal domain and an α-helical C-terminal domain, a typical fold for the thermolysin family of metallo-proteinases. The prometalloproteinase activation apparently occurs by cleavage of the Glu$^{208}$-to-Ala$^{209}$ peptide bond releasing a mature aureolysin with a calculated molecular mass of 33,306 Da (38-kDa by SDS-PAGE). Chromosomal DNA sequence information revealed a coding sequence of 1,527 nucleotides with the potential of encoding the aureolysin preproenzyme of 509 amino acids with a molecular mass of 56,321 Da. Little is known about the exact role of aureolysin in the pathogenicity of S. aureus. In vitro, aureolysin has been shown to cleave the plasma proteinase inhibitors, α$_2$-antichymotrypsin and α$_2$-proteinase inhibitor (Potempa, J et al.,1991; Potempa, J et al.,1986), and to activate prothrombin in human plasma (Wegrzynowicz, Z et al., 1990). It may also affect the stimulation of T and B lymphocytes by polyclonal activators and display inhibitory activity against immunoglobulin production by lymphocytes (Prokesova, L. et al., 1991). In addition, aureolysin activates the precursor of the glutamyl endopeptidase (V8 protease) secreted by this same microorganism (Drapeau, G. R. 1978).
Note that this data sheet is not lot-specific. Please consult the vial label and the certificate of analysis for information on specific lots.

4. References


