

JIS

JAPANESE INDUSTRIAL STANDARD

Methods for quantitative analysis of immobilized trypsin

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**In the event of any doubt arising,
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Errata will be provided upon request, please contact:

**Business Department,
Japanese Standards Association**
4-1-24, Akasaka, Minato-ku,
Tokyo, JAPAN 107
TEL. 03-3583-8002
FAX. 03-3583-0462

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Methods for quantitative analysis
of immobilized trypsin

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1. Scope This Japanese Industrial Standard specifies the methods for quantitative analysis to determine the quality of immobilized trypsin to be used for chemical products or for reagent.

Remarks 1. The standards cited in this Standard are listed in Attached Table 1.

2. The units and numerical values given in { } in this Standard are based on the traditional units and are appended for informative reference.

2. Definitions The definitions of terms used in this Standard shall follow JIS K 0211 and JIS K 3600.

3. Matters in common The general matters commonly used in chemical analysis shall follow JIS K 0050. Water shall be, however, A3 or A4 specified in JIS K 0557.

4. Classification of quantitative analyses The quantitative analyses of immobilized trypsin shall be classified as follows, depending on the kind of inhibiting materials which give peculiar inhibition on enzyme activity.

(1) Aminobenzamidine method React the low-molecular inhibiting material upon the part of protein where enzyme activity exists to make peculiarly adsorbing, wash out the inhibiting material which didn't make peculiarly adsorbing, nor reaction yet, and measure the absorbance shown by desorbed and separated inhibiting material, to determine the protein. This method can determine the immobilized trypsin from 0.35 to 3.5 mg.

Remarks: Low-molecular inhibiting materials are such as benzamidine and its derivatives, and leupeptin.

(2) Trypsin-inhibitor (soybean) method React the high-molecular inhibiting material upon the part of protein where enzyme activity exists to make peculiarly adsorbing, wash out the inhibiting material which didn't make peculiarly adsorbing nor reaction yet, and fractionally measure the desorbed and separated inhibiting material using high-performance liquid chromatography, to determine protein. This method can determine the immobilized trypsin of 0.35 mg.

Remarks: High-molecular inhibiting materials are such as trypsin inhibitor (soybean), trypsin inhibitor (albumen), and aprotinin.

5. Immobilized-trypsin suspension Immobilized-trypsin suspension shall be as follows.

5.1 Sample solution for analysis Control the concentration of sample immobilized trypsin with water so as to make it fall in the applicable range of each determination method.