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## Pepsin, Purified

—A white or yellowish-white powder, spongy mass, or translucent scales or granules. Freely soluble in water, producing more or less opalescence; practically insoluble in alcohol, in chloroform, and in ether. Purified Pepsin used in the second tier of the *Dissolution* test has an activity that is determined by the following method.

### Activity

**Pepsin solution:** Transfer about 2.5 mg of Purified Pepsin, accurately measured, to a 100-mL volumetric flask, dilute with 10 mM hydrochloric acid to volume, and mix.

[NOTE—Prepare immediately before use.]

**2.0% Hemoglobin solution:** Dissolve and dilute 2.5 g of bovine hemoglobin with water to 100 mL. Dilute 80 mL of this solution with 0.3 M hydrochloric acid to a volume of 100 mL.

**Trichloroacetic acid solution:** Dilute 5 g of trichloroacetic acid with water to 100 mL.

**Test Solution:** Transfer 5.0 mL of 2.0% *Hemoglobin solution* to a suitable container equilibrated at 37°. Add 1.0 mL of *Pepsin solution*, mix by swirling, and incubate at 37° for 10 minutes. Immediately add 10.0 mL of *Trichloroacetic acid solution*, mix by swirling, and incubate at 37° for 5 minutes. Pass through a filter having a 0.8- $\mu$ m or finer porosity.

**Control solution:** Transfer 5.0 mL of 2.0% *Hemoglobin solution* to a suitable container equilibrated at 37°. Mix by swirling, and incubate at 37° for 10 minutes. Immediately add 10.0 mL of *Trichloroacetic acid solution* and 1.0 mL of *Pepsin solution*, mix by swirling, and incubate at 37° for 5 minutes. Pass through a filter having a 0.8- $\mu$ m or finer porosity.

**Procedure:** Determine the absorbances of the *Test solution* and *Control solution*, in 1-cm cells, at a wavelength of about 280 nm, using water as the reference. Calculate the activity of the portion of Purified Pepsin taken by the formula:

$$10,000(A_U - A_C)$$

in which  $A_U$  and  $A_C$  are the absorbances of the *Test solution* and the *Control solution*, respectively.

**Auxiliary Information** - Please [check for your question in the FAQs](#) before contacting USP.

Topic/Question	Contact	Expert Committee
PEPSIN, PURIFIED	<a href="#">Margareth R.C. Marques</a> Principal Scientific Liaison	HDQ Headquarters

### Most Recently Appeared In:

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