

ProductInformation

HAPTOGLOBIN From Pooled Human Plasma

Product No. **H1511**
Store at 2-8°C

Product Description

Haptoglobin is found in normal plasma at a concentration of 30-190 mg/dl and accounts for 0.4-2.6% of the total plasma proteins. Some conditions such as cancer and coronary artery disease can raise this level, while diseases such as jaundice and cirrhosis can significantly lower the amount of haptoglobin in plasma. Human haptoglobin occurs naturally as 3 major phenotypes: Type 1-1, Type 2-1, and Type 2-2. The population distribution of phenotypes among Americans and Canadians is as follows:^{1,3,4}

Haptoglobin Type 1-1 - 17.4%
Haptoglobin Type 2-1 - 48.7%
Haptoglobin Type 2-2 - 33.9%

Mean g/L values for each phenotype in normal sera are:

Haptoglobin Type 1-1 - 1.77
Haptoglobin Type 2-1 - 1.40
Haptoglobin Type 2-2 - 1.06

Haptoglobin binds irreversibly with free native hemoglobin that has been released during hemolysis. This binding prevents iron loss and also prevents renal damage from the hemoglobin. The entire hemoglobin/haptoglobin complex is then metabolized by the body.⁴ The hemoglobin binding capacity of haptoglobin has been reported to be 1 mg hemoglobin bound for every 1.3 mg of haptoglobin.^{1,4} The complex also exhibits a peroxidase activity which can be used to monitor the binding, although the activity is not thought to be biologically significant.³

Haptoglobin's three common phenotypes, Type 1-1, Type 2-1, and Type 2-2 can be distinguished using linear gradient polyacrylamide gel electrophoresis (2.5-27%). Type 1-1 is a single band farthest from the origin, and is estimated to have a molecular weight of 100,000.³ Haptoglobin types 2-1 and 2-2 appear as a series of bands nearer to the origin. Types 2-1 and 2-2 have increasing molecular weights with some polymers being reported to be as high as 900,000.² Haptoglobin has also been reported to display other phenotypes that are thought to be significant in population studies.²

Haptoglobin is a glycoprotein having the following carbohydrate concentrations:⁵

Hexose	7.8%
(Galactose/mannose approx. 2:1)	
Acetylhexosamine	5.3%
Sialic acid	5.3%
Fucose	0.2%
Total carbohydrate	19.3%

At Sigma, haptoglobin phenotypes are prepared from human plasma by a modification of the method of Javid J. and Liang J.C., *J. Lab. Clin. Med.*, **82**, 991-1002 (1973). Each individual plasma unit is typed for haptoglobin phenotype and pooled with similar units before purification. Haptoglobin from pooled human plasma, Product No. H1511, is a mixture of all three phenotypes. This preparation retains its biological activity and has a minimum purity of 98% by agarose gel electrophoresis.

Precautions and Disclaimer

For laboratory use only. Not for drug, household or other uses. POTENTIAL BIOHAZARD. Handle as if capable of transmitting infectious agents.

Storage/Stability

Store at 2-8°C. Haptoglobin is supplied in serum vials containing 1 mg of haptoglobin per vial. Vials may be reconstituted with deionized water. Store reconstituted vials below 0°C.

Product Profile

Agarose gel: 1 μ l of a 10 mg/ml solution is applied to a 1% agarose gel. The buffer employed is 0.05 M sodium barbital and 0.01 M barbital, pH 8.6. The gel is electrophoresed at 120 volts for 1 hr and stained with 0.25% amido black.

Haptoglobin-binding Assay: The assay used is a variation of Nyman, Scand. J. Clin. Lab. Invest., **12**, 121-130 (1960). 10 mg/ml solutions of haptoglobin and hemoglobin are prepared. The haptoglobin solution is then divided into 100 μ l aliquots and the hemoglobin solution is added to each aliquot in 10 μ l graduations. The solutions are shaken at 37°C for 1/2 hr and then electrophoresed on cellulose acetate plates using a buffer containing 0.015 M boric acid, 0.14 M Tris, 0.004 M EDTA, pH 8.6. The plates are subjected to 180 volts for 20 min. The plates are stained with 0.2% dianisidine stain followed by a 0.2% Ponceau stain. When a hemoglobin (dianisidine) and haptoglobin (Ponceau S) band appear, the haptoglobin has been saturated.

One mg of haptoglobin will bind 0.5-0.9 mg hemoglobin.

References

1. Jayle and Morretti, Progress in Hematology, **3**, 342-359 (1962)
2. Laurell and Gronvall, Advances in Clinical Chemistry, **5**, 135-172 (1962)
3. Putnam, ~~The Plasma Proteins~~ Vol. II: 1-46 (1975)
4. Pintera, ~~Series Haematologica~~ Vol. IV, 2: 1-17 (1971)
5. Schultze and Heremans, ~~Molecular Biology of Human Proteins~~, Vol I: 200 (1966)

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