### **Technotes**

Blood coagulation and Anemia • Bone Metabolism • CARDIAC MARKERS • Fertility and Pregnancy • Hormone Markers • Immunology and Serology • Infectious Diseases • Inflammation • Kidney Diseases • Metabolic Syndrome • Neuroscience • Thyroid Diseases • Tumor Markers • Veterinary

# Myoglobin

Myoglobin is a small heme-containing protein (153 a.a.r., MW (w/o heme) 17053 Da and theoretical pI=7.29), that is responsible for the oxygen deposition in muscle tissues. Only one form of myoglobin is expressed in cardiac and skeletal muscles.

Myoglobin has been used as a marker of myocardial damage for almost six decades (1). It is commonly used in clinical practice as an early marker of AMI (2). However, due to the high concentration of myoglobin in skeletal muscle tissues, even minor skeletal muscle injury increases the myoglobin levels in blood (3). Therefore, myoglobin alone is not considered to be a reliable and sufficient marker in AMI diagnosis. Instead, it should be used together with cTnI or cTnT analysis as part of a multimarker strategy.

#### **HUMAN MYOGLOBIN**

Myoglobin is purified from human cardiac tissue by several chromatographic steps including gel-filtration and anion-exchange chromatography.

After SDS-PAGE in reducing conditions myoglobin is presented by a single band with apparent molecular mass of  $17~\mathrm{kDa}$  (Fig. 1).

CLINICAL UTILITY

Myocardial damage

Acute myocardial infarction

In 2001 Hytest's myoglobin preparation was selected by International Federation of Clinical Chemistry as an International Standard Material.

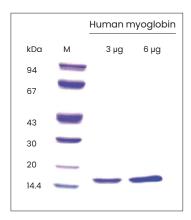


Figure 1.

SDS-PAGE of human myoglobin. M is the molecular weight standard (Pharmacia).

TECHNOTES • MYOGLOBIN

## MONOCLONAL ANTIBODIES SPECIFIC TO MYOGLOBIN

Hybridoma clones have been derived from hybridization of Sp2/0 myeloma cells with spleen cells of Balb/c mice immunized with purified human myoglobin. Different combinations of monoclonal antibodies can be used for the immunoassay development. The best MAb combinations for sandwich immunoassay are:

**4E2cc** – **7C3cc** 7C3cc – 4E2cc

#### **MYOGLOBIN FREE SERUM**

Myoglobin free serum is prepared from pooled normal human serum by immunoaffinity chromatography. The matrix for affinity sorbent utilizes several monoclonal antibodies with different epitope specificity. Based on ELISA testing the level of myoglobin in myoglobin free serum is below 0.1 ng/ml (Fig. 2).

Myoglobin free serum is used as a matrix for standard and calibrator preparations.

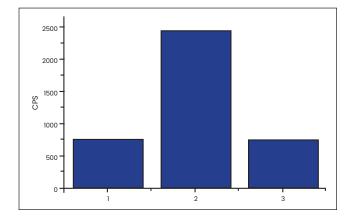


Figure 2.

Myoglobin level in normal human serum and in myoglobin free serum detected in sandwich-immunoassay. Column 1: Buffer, column 2: Pooled normal human serum before myoglobin extraction (4 ng/ml of myoglobin) and column 3: Residual signal in myoglobin free serum.

#### **REFERENCES**

- Kiss A and Reinhart W. (1956) Determination of myoglobin in serum and in urine after heart infarction. Wien Klin. Wochenschr. 68(9):154– 155.
- 2. Penttila K. et al. (2002) Myoglobin, creatine kinase MB isoforms and creatine kinase MB mass in early diagnosis of myocardial infarction in patients with acute chest pain. Clin Biochem. 35(8), 647-653.
- 3. Van Nieuwenhoven FA, et al. (1995) Discrimination between myocardial and skeletal muscle injury by assessment of the plasma ratio of myoglobin over fatty acid-binding protein. Circulation. 92(10), 2848-2854.

#### ORDERING INFORMATION

#### **MONOCLONAL ANTIBODIES**

Product name	Cat.#	MAb	Subclass	Remarks
Myoglobin	4M23	4E2cc	IgG1	In vitro, EIA
		7C3cc	IgG1	In vitro, EIA
		1B4	IgG1	EIA

#### ANTIGEN

Product name	Cat.#	Purity	Source
Myoglobin	8M50	>95%	Human cardiac muscle

#### **DEPLETED SERUM**

Product name	Cat.#	Source
Myoglobin free serum	8MFS	Pooled normal human serum

Please note that some or all data presented in this TechNotes has been prepared using MAbs produced *in vivo*. MAbs produced *in vitro* are expected to have similar performance.

