BIOTECH SUPPORT GROUP

# HemogloBind™

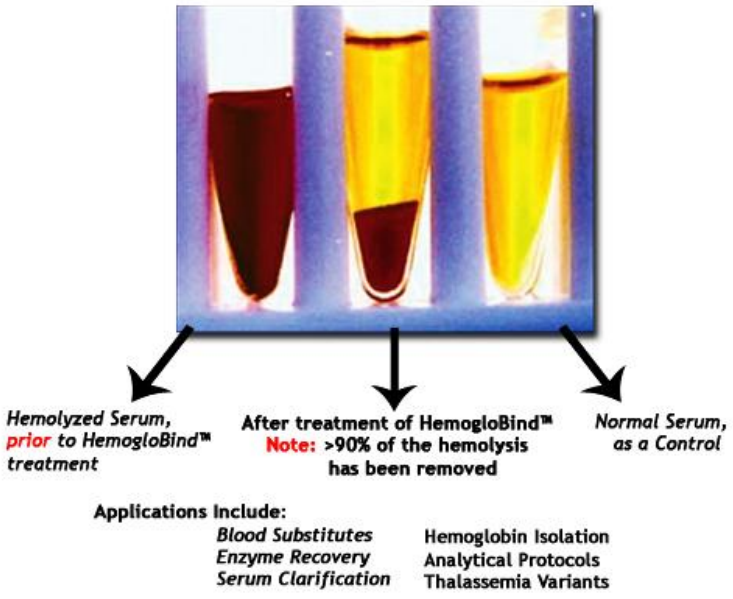
**HemogloBind™ Hemoglobin Removal and Capture**

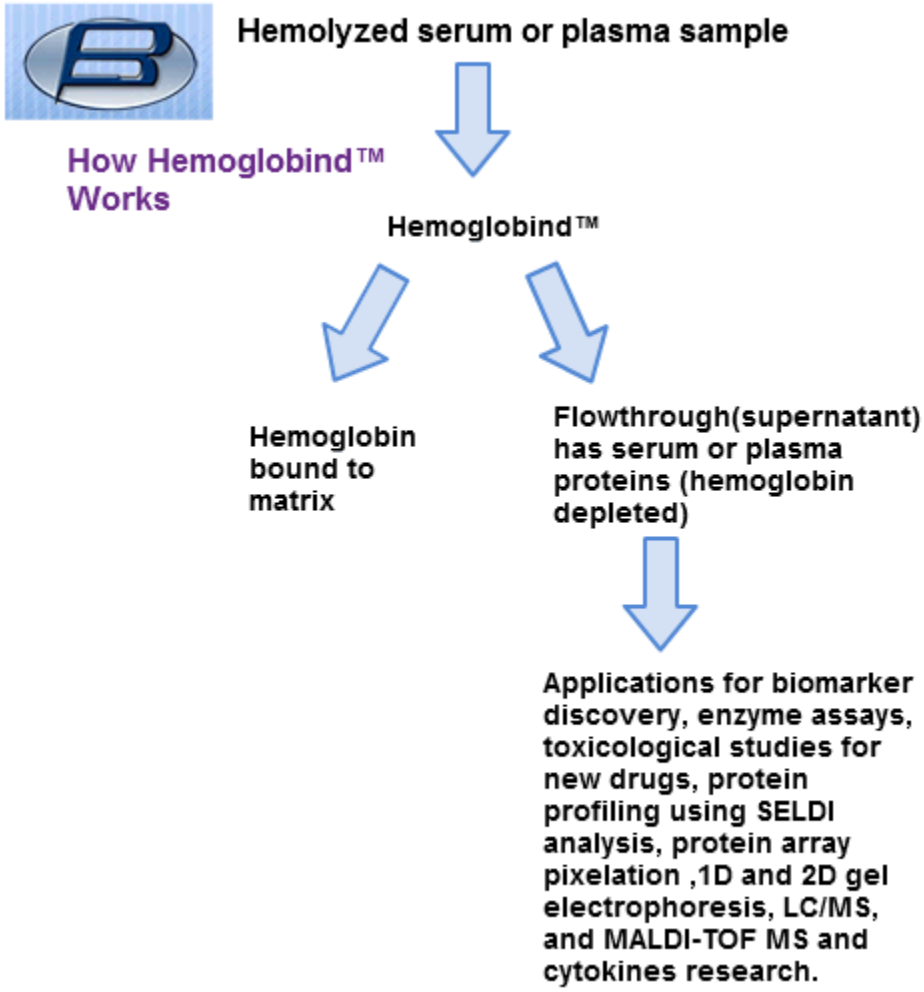
- Has a high degree of specificity for hemoglobin
- Suitable for serum/plasma or red cell lysates without cross-reacting with other analytes[1,3,4]
- Distinguishes between hemoglobin variants
- Applications in blood substitutes, enzyme recovery and analytical interferences[1,2]

Poly-electrolytes are polymers with repeating units of stationary charges. **HemogloBind™** comes from a class of solid-phase, or surface-based, elastomeric poly-electrolytic surfaces that bind proteins through an empirically derived chemistry combining elements of polymer composition, cross-linking architecture and charge properties. As with bio-polymers like DNA and Heparin, governing their reactivity is the spatial presentation of the electrostatic groups along a flexible polymer chain. This strategy was used in the creation of both Viraffinity™ and **HemogloBind™**.

**HemogloBind™** is engineered for a high degree of selectivity and does not cross react with most common serum components, making it an excellent tool in numerous applications. These include analytical protocols where optical interference is problematic, such as bilirubin analysis and bulk serum clarification. Hemoglobin variants, as in thalassemia, bind with differential affinity towards **HemogloBind™**.

For purification and/or analysis of hemoglobin, a modest elevation in pH will facilitate desorption of hemoglobin bound to **HemogloBind™**.






| Product | Size | Item No. | Price |
|---|---|---|---|
| **HemogloBind™** | 15ml | HO145-15 | $355 |
| **HemogloBind™** | 50ml | HO145-50 | $775 |

## PROTOCOL

Supplied as an aqueous suspension of synthetic, pH 6.5. The reagent when not used must be kept sealed and stored at 4°C. Do not freeze. HemogloBind™ retains full activity when stored at 4°C for 6 months. Expiration date is shown on product label.

**To treat 250 µl of hemolyzed serum using SPIN-X Tubes (Recommended):**

1. Shake the HemogloBind™ suspension.
2. Using wide-bore pipette tips, pipette 250 µl or 500ul of the HemogloBind™ suspension into the small tub of the SPIN-X set.
3. Add 250 µl of the hemolyzed serum to the small tub. (~10 mg/mL Hb)
4. Vortex for 20 seconds.
5. Mix by inversion for 15 minutes.

6. Centrifuge for 1-2 minutes at 9000 RPMs.

Supernatent contains hemoglobin depleted sample, while the solid contains the hemoglobin removed.

**To treat 250 µl of hemolyzed serum using microfuge tubes:**

1. Shake the HemogloBind™ suspension.
2. Using wide-bore pipette tips, pipette 250 µl or 500ul of the HemogloBind™ suspension.
3. Add 250 µl of the hemolyzed serum. (~10 mg/mL Hb)
4. Vortex for 30 seconds.
5. Mix by inversion for 15 minutes.
6. Centrifuge for 1-2 minutes at 9000 RPMs.

Supernatent contains hemoglobin depleted sample, while the solid contains the hemoglobin removed.

Other Analytes

HemogloBind™ cannot reduce interference caused by substances released from erythrocyte hemolysis other than that caused by hemoglobin. It is compatible with Bilirubin, Total Protein, Immunoglobulin, Albumin, Creatinine, ALT, AST, GGT, Creatine Kinase, LDH, BUN, Amylase, Cholinesterase. It is marginally compatible with Alkaline Phosphatase. It is not compatible with Calcium, Magnesium.

Haptoglobin (HAP) Influence

The extent of hemoglobin removal may be influenced by the presence of elevated haptoglobin concentrations and sialo-glycoprotein which may be present in some acute-phase adult serum samples.

Myoglobin Binding

HemogloBind will not bind to Myoglobin, a protein that is structurally similar to hemoglobin but of lower molecular mass.

Hemoglobin Variants

Hemoglobin variants, as in thalassemia, and glycosylated hemoglobin bind with differential affinity towards HemogloBind™.

Desorption of Bound Hemoglobin

For purification and/or analysis of hemoglobin, 100 mM Tris-Borate, pH 9, will facilitate desorption of hemoglobin bound to HemogloBind™.

# References

**Biological Fluids**

J Krupey - United States Patent: 10/180,053, 2002 Removal of extraneous substances from biological fluids containing nucleic acids and the recovery of nucleic acids

**Red Cell Lysates**
Kyoungsook Park, Christopher D. Saudek, and Gerald W. Hart Increased Expression of β-N-Acetylglucosamindase (O-GlcNAcase) in Erythrocytes from Prediabetic and Diabetic Individuals.Diabetes.2010;59(7):1845-50.

**Stored Blood Products**
Delobel J., Rubin O., Prudent M., Crettaz D., Tissot J.-D., Lion N.(2010) Biomarker Analysis of Stored Blood Products: Emphasis on Pre-Analytical Issues. International Journal of Molecular Sciences. 11(11):4601-4617

**Red Blood Cells**
Alvarez-Llamas, G., de la Cuesta, F., Barderas, M. G., Darde, V. M., Zubiri, I., Caramelo, C., Vivanco, F. A novel methodology for the analysis of membrane and cytosolic sub-proteomes of erythrocytes by 2-DE.Electrophoresis.2009;30:4095-4108

Zihao Wang, Kyoungsook Park, Frank Comer1, Linda C. Hsieh-Wilson, Christopher D. Saudek, Gerald W. Hart. Site-Specific GlcNAcylation of Human Erythrocyte Proteins: Potential Biomarker(s) for Diabetes Mellitus. Diabetes.2008;58, 309-317.

Datta, Pradip. Effect of Hemolysis, High Bilirubin, Lipemia, Paraproteins, and System Factors on Therapeutic Drug Monitoring. Handbook of Drug Monitoring Methods.2008; 97-109.

Yuichi Miki, Tomoki Tazawa, Kazuya Hirano, Hideki Matsushima, Shoko Kumamoto, Naotaka Hamasaki, Tomohiro Yamaguchi, Masatoshi Beppu. Clearance of oxidized erythrocytes by macrophages: Involvement of caspases in the generation of clearance signal at band 3 glycoprotein. Biochemical and Biophysical Research Communications.2007; 363(1):57-62

Sarawathi,et al., Relative quantification of glycated Cu-Zn superoxide dismutase in erythrocytes by electrospray ionization mass spectrometry, Biochimica et Biophysica Acta. 1999.1426(3):483-90

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Person, N.B., Effect Of HemogloBind™ On Interference Reduction In Bilirubin Analysis.poster Clinichem, 1995.

**Serum**
Baion, C.M. & Ali, A.C.Evaluation Of HemogloBind™ For Removal Of O-Raffinose Crosslinked Hemoglobin (Hemolink™) From Serum, poster AACC Meeting 1997.

**Tissue**
Padilla, S., Convenient Method for Decreasing the Amount of Hemoglobin in Tissue Samples Without Affecting the Level of Cholinesterase Activity, unpublished personal correspondence, 1994.

## CONTACT US

We welcome your questions, comments and concerns regarding our products.

BIOTECH SUPPORT GROUP


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