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HemoVoid[™] Blood Card Reagent Hemoglobin Depletion And Protein Enrichment From

Dried Whole Blood Cards

- Hemoglobin voids in flow-through >98%, with <30 minute bind/wash/elute protocol
- · Hemoglobin removal from whole blood lysates extracted from dried blood cards
- Blood proteins and enzymes are enriched for potential biomarker and proteomic studies.
- Hemoglobin removal from frozen and fresh whole blood.
- Removes hemoglobin from diverse species incl. human, sheep, bovine, goat, rat, etc.

Hemoglobin is a common contaminant from dried whole blood cards and not normally found in serum samples. The HemoVoid[™] Blood Card protocol was designed to substantially reduce the presence of hemoglobin and its associated interference with many serum protein analytes.

HemoVoid[™], a silica-based mixed mode matrix, removes hemoglobin from dried whole blood card samples. The HemoVoid[™] protocol uses mild buffers; the protocol conditions are so gentle that native enzyme activity is retained in elution fractions.







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SDS Page (4-20%) Sheep Blood(left)

Mouse blood (right)

Product	# of samples processed	Item No.	
HemoVoid™ Blood Card	10 Dried Whole Blood Card 0.5" Spots	HVBC-10	
HemoVoid™ Blood Card	50 Dried Whole Blood Card 0.5" Spots	HVBC-50	
NOTE: Please contact sales@biotechsupportgroup.com for prices in bulk amount.			

Kit Content	10 Prep	50 Prep	Reagent
HemoVoid™	0.5 gram	2.5 grams	Supplied
Protein Extraction Buffer PEB	5 ml	25 ml	Supplied
Binding Buffer HVBB, PH 6.0	15 ml	75 ml	Supplied
Wash Buffer HVWB, PH 7.0	15 ml	75 ml	Supplied
Elution Buffer HVEB, PH 9.8	3 ml	15 ml	Supplied
SpinX Centrifuge tube filters	10	50	Supplied
Suggested Or Equivalent Supplier of Blood Card: Whatman 903 [™] Protein Saver cards			Not Supplied

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HemoVoid[™] Protocol For Hemoglobin Depletion From Blood Spot/Blood Card

Based on processing 20-50 µl whole blood applied to and dried on Whatman 903™ Protein Saver cards (approximately equivalent to the imprinted 0.5" circle)

- Extraction of dried protein from the card. Punch out the dried blood section from the card into a microfuge tube. Add 400 μl PEB buffer. Shake for 30 minutes at room temperature. Microfuge at 5000 rpm for 4 minutes. Transfer the protein sample to microtube.
- 2. Weigh out 50 mg of $\textbf{HemoVoid}^{\texttt{m}}$ matrix into the supplied SpinX filter.
- 3. Add 400 µl of **Binding Buffer HVBB** to the SpinX filter. Vortex or mix well for 5 minutes at room temperature followed by centrifugation at 3000 rpm. Discard the supernatant.
- 4. Repeat step 3.
- 5. Add 300 µl of **Binding Buffer HVBB** to the SpinX filter. Add 300 µl of the Sample prepared in step 1 to the same SpinX filter. Vortex for 10 min and then centrifuge for 2 minutes at 5000 rpm.
- 6. Discard the hemoglobin containing filtrate.
- 7. To the pellet, add 500 µl of **Wash Buffer HVWB**. Vortex or mix well for 5 min and centrifuge for 2 minutes at 5000 rpm. Discard the filtrate.
- 8. Repeat Step 7, twice.
- 9. To the pellet, add 200 µl of **Elution Buffer HVEB**. Vortex or mix well for 10 min and centrifuge for 2 minutes at 5000 rpm. Analyze the hemoglobin depleted elute protein.

Related HemoVoid™ References

Human Red Blood Cells (RBC)

HemoVoid[™] On Bead Digestion Application Work On RBC by Irene Granlund, Umeå University

Red Blood Cells, Plasmodium extracts

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P. Falciparum Clone 3D7 Cultured In Human Erythrocytes

Lasonder E, Green JL, Camarda G, Talabani H, Holder AA, Langsley G, Alano P. <u>The Plasmodium</u> <u>falciparum schizont phospho-proteome reveals extensive phosphatidylinositol and cAMP-Protein Kinase A</u> <u>signalling</u>. J Proteome Research. 2012;

Red Blood Cell Lysate

Barasa, Benjamin, and Monique Slijper. "<u>Challenges for red blood cell biomarker discovery through</u> <u>proteomics</u>." *Biochimica et Biophysica Acta (BBA)-Proteins and Proteomics* 1844.5 (2014): 1003-1010.

Lange, Philipp F., Pitter F. Huesgen, Karen Nguyen, and Christopher M. Overall. "<u>Annotating N termini for</u> <u>the Human Proteome Project: N termini and Na-acetylation status differentiate stable cleaved protein</u> <u>species from degradation remnants in the human erythrocyte proteome</u>." *Journal of proteome research* (2014).

Katja Walpurgis, Maxie Kohler, Andreas Thomas et al.<u>Validated hemoglobin-depletion approach for red</u> blood cell lysate proteome analysis by means of 2D-PAGE and Orbitrap MS.Electrophoresis.2012;

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Sudha Neelam, David G Kakhniashvili, Stephan Wilkens et al. <u>Functional 20S proteasomes in mature</u> <u>human red blood cells</u> Experimental Biology and Medicine.2011;236:580-591

CONTACT US

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