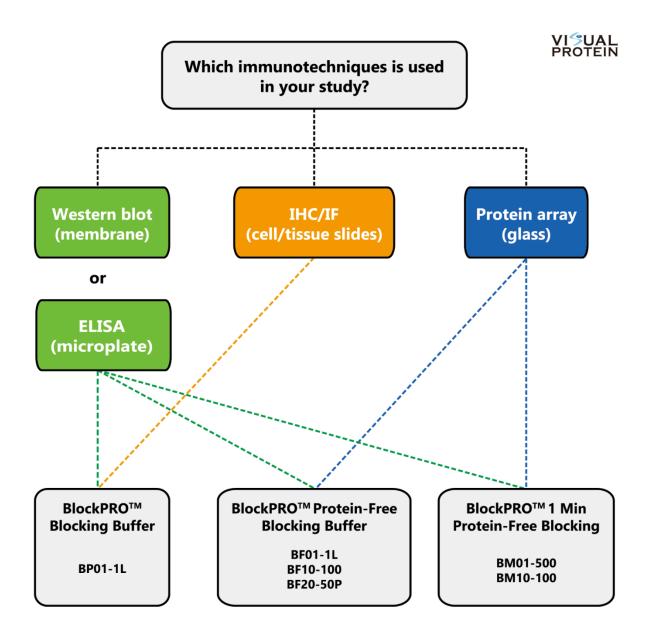


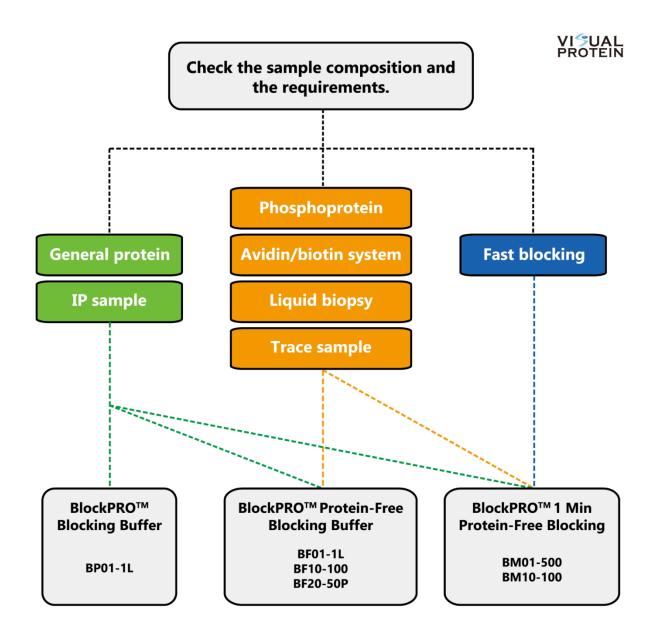
BlockPRO™ Series Blocking Buffer

- 1. BlockPRO™ Blocking Buffer
- 2. BlockPRO™ Protein-Free Blocking Buffer
- 3. BlockPRO™ 1 Min Protein-Free Blocking Buffer

Before probing for proteins of interest, the remaining binding surface of the membrane must be blocked to prevent the nonspecific binding of the antibodies. Otherwise, the antibodies or other detection reagents will bind to any remaining sites on the membrane that initially served to immobilize the proteins of interest. In principle, any protein that does not have binding affinity for the target or probe components in the assay can be used for blocking.

The ideal blocking buffer will bind to all potential sites of nonspecific interaction, eliminating background altogether without altering or obscuring the epitope for antibody binding. Blocking buffers can influence antibody binding and specificity- so optimization is needed. No single protein or mixture of proteins works best for all Western blot experiments, and empirical testing is necessary to obtain the best possible results for a given combination of specific antibodies, membrane type and substrate system.





We offer three types of blocking buffer for optimizing the detection of the target proteins.

	Product name	BlockPRO Blocking Buffer	BlockPRO Protein-Free Blocking Buffer	BlockPRO 1 Min Protein-Free Blcoking Buffer
	1×100 ml (×10)	_	BF10-100	BM10-100
Size	1×500 ml	_	-	BM01-500
	2×500 ml	BP01-1L	BF01-1L	-
	20x2.5 ml (x20)	-	BF20-50P	-
Blocking system		Single purified protein	Chemical base (protein free)	Chemical base (protein free)
Blocking incubatin time		30 to 60 minutes	30 to 60 minutes	1 to 3 minutes
Base buffer		TBS Buffer	TBS Buffer	TBS Buffer
Tween-20		0.05%	0.05%	0.05%
Increase of signal intensitiy		•	•	•
Enhance of specific signal		•	•	•
Reduction of background noise		•	•	•
Ready-to-use		BP01-1L	BF01-1L	BM01-500
Safeness		DI 01 12	B. 01 11	Bivior 300
Lot to lot consistency		•	•	•
Washing	PBST			•
buffer	TBST			
Dullel	Western blot (PVDF)		•	
Application	Western blot (NC)	+		
	ELISA	+		
	IHC	<u> </u>		
	Antibody diluent			
Compatibility	Phosphoprotein detection	-		
	Avidin/Biotin system			
Reference	Aviditi/ bioditi system			
Official Link		https://www.visualprotein.com/en/product/8/10/9	https://www.visualprotein.com/en/product/8/10/30	https://www.visualprotein.com/en/product/8/10/32
Highlights		Performs well with a wide range of antibodies and antibody combinations Single purified protein provides fewer chances of cross-reaction with assay components than serum or milk solutions Blocks in less than 60 minutes Works with both nitrocellulose and low fluorescence PVDF	reactivity associated with protein- based blocking buffers. 2. Sample-and-antibody combinations require the elimination of all possible exogenous animal proteins in the assay system to avoid cross-	Enhanced formula of protein-free blocking bufffer Sample-and-antibody combinations require the elimination of all possible exogenous animal proteins in the assay system to avoid cross-reaction or quenching of the desired probe function Shorten the blocking time to less than 3 minutes
When to use		Best for med-high abundant proteins or strong antibody affinity High background with current blocking buffer Stripping and reprobing western blots	1. For low abundant target protein or poor immunoreactivity antibodies 2. Use when protein-based blockers cause high background 3. Use when targeting phosphoproteins 4. Best to use when storing reused antibodies in blocker 5. Use as antibody diluent 6. Use as storing solution to saving antibodies	1. When time is essensital 2. Use when protein-based blockers cause high background 3. Use when targeting phosphoproteins 4. Best to use when storing resused antibodies in blocker 5. Use as antibody diluent 6. Use us storing solution to saving antibodies

BlockPRO™ Blocking Buffer

BlockPRO™ Blocking Buffer is based on single purified protein which is suitable for blocking in Western blot, ELISA, immunohistochemistry and other immunochemical application. It can block excess binding site but not cover on the binding protein and therefore increase the signal intensity. Best for med-high abundant proteins or strong antibody affinity; high background with current blocking buffer; stripping and re-probing Western blots. Present better data result than milk and BSA blocking buffer.

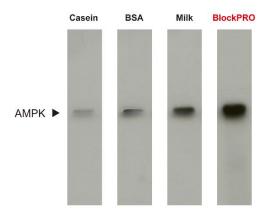


Figure 1. Signal strength comparison of Casein, BSA, milk, and BlockPRO™ Blocking Buffer.

Loading 30 µg cell lysate (HepaG2) and detect with anti-AMPK (mouse, 1:1,000). Secondary antibody: antimouse IgG-HRP 1:10,000. Membrane: Hyond™ P. Detection: Hyperfilm™ ECL. All results were exposed for 30 seconds and capture by X-ray film.

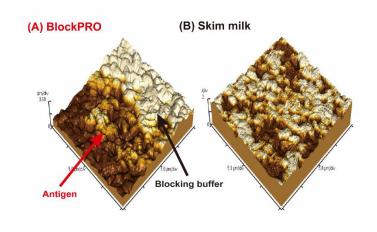


Figure 2. Blocking with skim milk or BlockPRO™ Blocking Buffer and detect by SEM.

30 µg cell lysate (HepaG2) separated by 12.5% SDS-PAGE and blocking by skim milk or BlockPRO™ Blocking Buffer. Membranes detected by SEM. The SEM result showed that BlockPRO™ Blocking Buffer only blocking on excess binding site and reveal the position of antigen and therefore will not reduce the signal intensity.

Website: www.visualprotein.com/en/product/8/10/9

BlockPRO™ Protein-Free Blocking Buffer

BlockPRO™ Protein Free Blocking Buffer is a non-protein formulation which enhances sensitivity and minimizes background noise, presenting better results than traditional protein-based blocking buffer in immunoassays. The synthetic formulation of BlockPRO™ Protein-Free Blocking Buffer makes it suitable for PVDF and nitrocellulose platform, avidin/biotin system, detection of phosphoprotein, and other immunochemical applications.

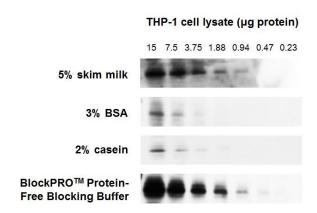


Figure 1. BlockPRO™ Protein-Free Blocking Buffer is better than protein-based blocking buffers (skim milk, BSA and casein) for detection of target protein in Western blotting.

THP-1 cell lysates were prepared and separated by electrophoresis. The proteins were transferred to PVDF and blocked for 1 hour at room temperature with the indicated blocking buffer, probed with mouse anti-pAMPK followed by anti-mouse HRP and detected by chemiluminescence. All results were exposed to X-ray film for 30 seconds.

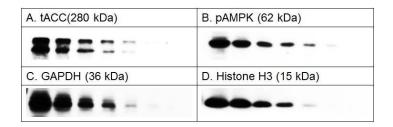


Figure 2. BlockPRO™ Protein-Free is suitable for various protein antigen detection, such as high molecular weight protein, tACC; phosphoprotein, pAMPK; abundant protein, GAPDH; low molecular weight protein, histone H3.

THP-1 cell lysates were prepared and separated by electrophoresis. The proteins were transferred to PVDF and blocked for 1 hour at room temperature with BlockPRO™ Protein-Free Blocking Buffer. Antibodies designed to probe the indicated proteins were used. All the signals were detected by chemiluminescence and were exposed to X-ray film.

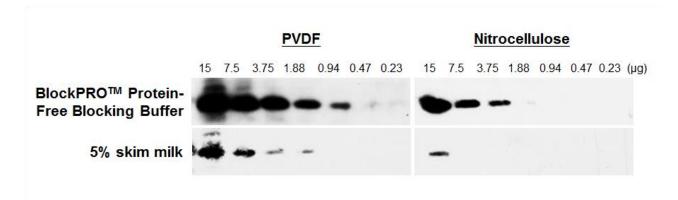


Figure 3. BlockPRO™ Protein-Free Blocking Buffer can be used in both PVDF and nitrocellulose platform. Hela cell lysates were prepared and separated by SDS-PAGE. The proteins were transferred to PVDF or nitrocellulose membranes. The membranes were blocked for overnight at 4 °C with BlockPRO™ Protein-Free Blocking Buffer or 5% skim milk, probed with mouse anti-histone H3 followed by anti-mouse HRP and detected by chemiluminescence. All results were exposed to X-ray film for 30 seconds.

Website: www.visualprotein.com/en/product/8/10/30

BlockPRO™ 1 Min Protein-Free Blocking Buffer

BlockPRO™ 1 Min Protein-Free Blocking Buffer is a fast blocking buffer for Western blotting when time is essential. It effectively prevents protein cross-reaction and improves the signal/noise ratio of the detected signals by one-minute blocking. BlockPRO™ 1 Min Protein-Free Blocking Buffer can be applied to immunodetection systems including antibodies and biotin/avidin systems such as ELISA, Western blotting, immunohistochemistry and other immunochemical applications.



Figure 1. BlockPRO™ 1 Min Protein-Free Blocking Buffer enhanced the signal intensity and shorten the blocking time to 1 min.

20 µg of Huh-7 cell lysates were serial diluted and separated by electrophoresis. The proteins were transferred to PVDF and blocked for 1 min hour at room temperature with BlockPRO™ 1 Min or 1 hour in competitor's product (as indicated), probed with mouse anti-ACC followed by anti-mouse HRP and detected with LumiFlash™ Ultima Chemiluminescent Substrate.

Website: www.visualprotein.com/en/product/8/10/32