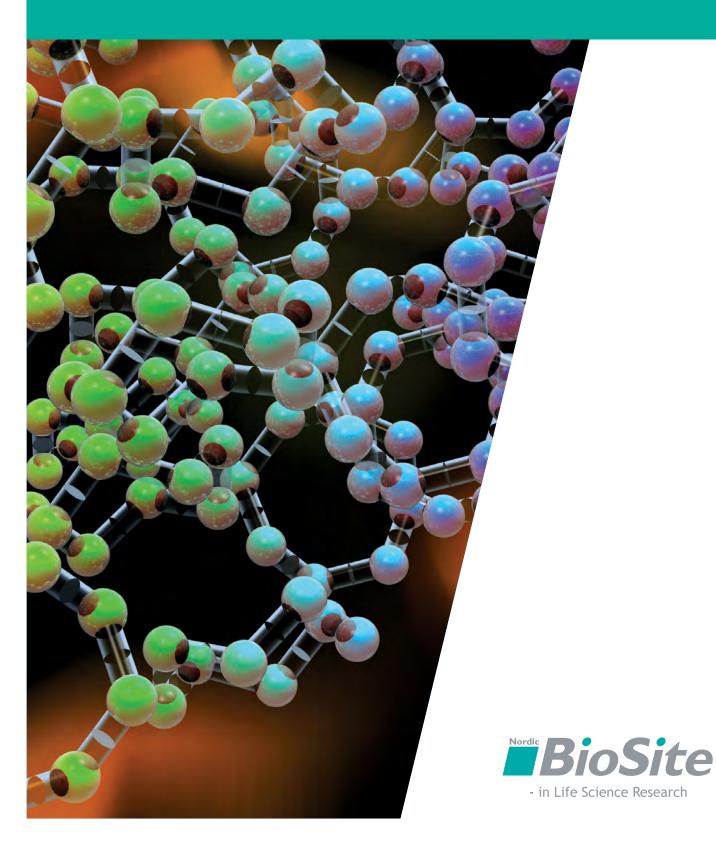


# Western Blot



# Ordering

Orders may be placed by any of the following methods:

Web:	www.nordicbiosite.com	
E-Mail:	order@nordicbiosite.com	
Phone:	+46 (0)8 5444 33 40	
Fax:	+46 (0)8 756 94 90	

# Western Blot

The Western blot (sometimes called the protein immunoblot) is a widely used analytical technique used to detect specific proteins in a sample of tissue homogenate or extract. It uses gel electrophoresis to separate native proteins by 3-D structure or denatured proteins by the length of the polypeptide. The proteins are then transferred to a membrane (typically nitrocellulose or PVDF), where they are stained with antibodies specific to the target protein. The gel electrophoresis step is included in Western blot analysis to resolve the issue of the cross-reactivity of antibodies.

Other related techniques include dot blot analysis, immunohistochemistry and immunocytochemistry where antibodies are used to detect proteins in tissues and cells by immunostaining, and enzyme-linked immunosorbent assay (ELISA).

The method originated in the laboratory of Harry Towbin at the Friedrich Miescher Institute. The name western blot was given to the technique by W. Neal Burnette[4] and is a play on the name Southern blot, a technique for DNA detection developed earlier by Edwin Southern. Detection of RNA is termed northern blot and was developed by George Stark at Stanford.

Nordic BioSite wants to facilitate your research and here you find reagents to get the perfect Western Blots.

Never hesitate to contact us if you have any questions about the products we offer. We are here to help you.

# Get optimal Western Blots with reagents from Nordic BioSite

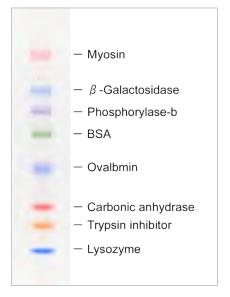
## **Protein Size Markers**

0,5 ml protein marker, Sizes: 70, 57, 40, 28, 18, 13.5, 8.5 kDa, cat # PAT-80189-1 0,5 ml protein marker, Sizes: 150, 100, 70, 50, 35, 25, 20, 15 kDa, cat # PAT-80190-1 DynaMarker® Protein MultiColor Stable cat# 181-DM660S

DynaMarker Protein MultiColor consist of eight proteins, prestained orange, blue, purple, green or red, ranging from approximately 15 kDa to 230 kDa The marker is stable at 4°C for as long as one year by the improved colorilization method.

Broad-Range Protein Marker-2 consists of eight precisely sized recombinant proteins (M.W. 150, 100, 70, 50, 35, 25, 20, 15 KDa). The concentration of each protein is about 0.1  $\mu$ g/ $\mu$ l

Mid-Range Protein Marker-1 consists of Seven precisely sized recombinant proteins (M.W. 70, 57, 40, 28, 18, 13.5, 8.5KDa). The concentration of each protein is 0.1  $\mu$ g/ $\mu$ l except for 8.5KDa (0.2  $\mu$ g/ $\mu$ l).



# **Control Protein**

Use our recombinant proteins for positive controls. Have a look in our catalog "BioSite Proteins" or search at www.nordicbiosite.com.

## Lysates

Our lysates includes organ specific lysates from various species. Pure cell line lysates and lysates from cells overexpressing certain proteins and many more alternatives. Search for relevant products among our >10 000 cell lysates at www.nordicbiosite.com.

## **Protein Determination**

#### **BCA Protein Assay**

The BCA Protein Assay is a detergent-compatible formulation based on bicinchoninic acid (BCA) for the detection and quantization of total protein.

Name	Size	Range	Cat #
BCA Protein Assay Kit	50 test tubes or 500 in 96-well format	20 to 2 000 ug/mL	KBB-264
Micro BCA Protein Assay Kit	100 test tubes or 500 in 96-well format	0.5 to 20 ug/mL	KBB-265

#### **Bradford Assay**

This protein assay, based on the method of Bradford, is a simple and accurate procedure for determining protein concentration. Protein extraction and sample dilution buffer are included in the kit to avoid interactions with buffer and detergent components.

Name	Size	Cat #
MaxDiscovery™ Bradford Assay Kit	1000 in 96-well format	228-3440-01

#### Gel staining

Coomassie staining is the most commonly used method to visualize protein bands in the gel after electrophoresis and does not chemically modify the target bands. This allows the visualization of the proteins in the gel as blue bands. The product can also be used to check the efficiency of the transfer step in Western Blot.

Silver staining is the choice for visualizing low abundant proteins since it it approximately 50 times more sensitive than Coomassie.

Name	Size	Cat #
Coomassie Blue Staining/ Destaining Solution	100 ml Staining Solution 100 ml Destaining solution For 10-20 assays	RBB-100
Coomassie Blue Fast Staining Solution	100 ml Staining Solution For 10- 20 assays	RBB-105
Silver staining kit	For 10 assays	RBB-102

#### Membranes

We offer 4 different membrane types:

Membrane Type	Pore Size µm	Insulin Binding	BSA Binding
PVDF 0.45 μm	0.45	Ca. 160 ug/cm2	Ca 215 ug/cm2
PVDF 0.2 µm	0.2	Ca. 262 ug/cm2	Ca 349 ug/cm2
PVDF Fluor	0.45	Ca 155 ug/cm2	Ca 205 ug/cm2
Nitrocellulose	0.45	Ca 80 ug/cm2	Ca 100 ug/cm2

The membranes are pre-cut to perfectly fit the below described incubation containers in order to optimize the amount of antibody solution needed and thereby save costs for antibodies.

Membranes are available in the following sizes:

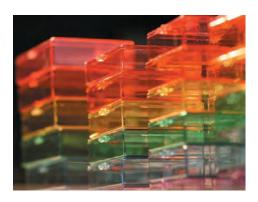
- Medium (8.3 x 6 cm size)
- Mini Square (8.3 x 7.3 cm size)
- Extra Large (13.5 x 8.5 cm size)

All membranes are available in packets of 10, 25 or 50.

# PerfectWestern Containers

Containers for all Western blotting steps: Antibody incubation, washing, & staining **35 different sizes in clear, black & transparent colors** For whole blots, half blots, strips, 2D gels, etc.

- Ultra-smooth flat inner surfaces
- Lids to minimize evaporation and fume exposure
- 90 edges for excellent membrane movement
- Stable up to 70 C for stripping, etc.



The most popular containers are listed below. Please contact Nordic BioSite or check online for other sizes.

Rectangular	Dimensions	Description	Cat #
		Clear	B109
PerfectWestern, Extra Large. For larger protein Gels	15,4 x 10, 4 x 3,6 cm	Black	B110
		Rainbow Set	B109S
		Clear	B101
PerfectWestern, Medium. For standard Mini-Gels	9,1 x 6,6 x 2,9 cm	Black	B103
		Rainbow Set	B101S
PerfectWestern, Mini Square.	8,6 x 8,6 x 3,1	Clear	B144
For larger Mini-Gels		Black	B145
		Clear	B104
PerfectWestern, Small. For Mini-Gels cut in half	7.3 x5.1 x 3.3	Black	B105
		Rainbow Set	B104S
PerfectWestern-Dura, Medium	9.1 x 6.6 x 3.2 cm	Flexible	B101-D

Sectional	Dimensions, outer	Well Dimensions	Cat #
PerfectWestern, 6-Sectional- Short, Clear/Black	17,3 x 8,4 x 2,7 cm	8,4 x 2,7 x 3,2 cm	B130 / B131
PerfectWestern, 6-Sectional- Long, Clear/Black	26,7 x 15,8 x 4,4 cm	15,8 x 4 x 4,4 cm	B134 / B135
PerfectWestern, 3-Sectional- Medium, Clear/Black	22,2 x 10 x 3,3 cm	10 x 6,6 x 3.3 cm	B101-3S / B103-3S

# **Primary Antibodies**

Nordic BioSite offers a very large selection of primary antibodies for Western Blotting. Search at www.nordicbiosite.com or contact us for help in finding the right antibody.



We also provide customized production of monoclonal and polyclonal antibodies.

# Protein Loading control Antibodies (House-keeping-proteins)

Genes that are constitutively and stably expressed at high levels in almost all tissues and cells are often referred to as housekeeping genes. Housekeeping proteins such as GAPDH, beta Actin, beta Tubulin, or Histone H2B (nuclear fractions) are useful as loading controls for western blots and protein normalization. Increasingly, researchers are making use of specific antibodies to housekeeping proteins to standardize and compare levels of protein. However, studies examining gene transcript and protein levels have shown that expression levels of housekeeping genes can vary. Collectively the results indicate that some housekeeping proteins may be better suited than others for use as loading controls depending on the model system used.

#### Unconjugated

Beta-Actin, (mouse monoclonal), cat # 60008-1-Ig Beta-Tubulin, (rabbit polyclonal), cat # 10068-1-AP GAPDH, (mouse monoclonal), cat # 60004-1-Ig

#### **Direct Conjugated**

HRP-conjugated Beta-Actin Antibody (mouse monoclonal), cat# HRP-60008

HRP-conjugated GAPDH Antibody (mouse monoclonal), cat# HRP-60004

All the antibodies above are reactive towards at least human, mouse and rat protein. More options are available; please contact Nordic BioSite for other suggestions.

# Secondary Antibody

You will need a secondary antibody for visualization if your primary antibody is not directly labeled. For Western Blotting the most common label on the secondary antibody is an enzyme like Horse Radish Peroxidise (HRP) for Enhanced Chemi-Luminiscence (ECL) or a fluorochrome.

Nordic BioSite offers a wide selection of secondary antibodies covering a broad selection of immunoglobulins from various species and cross absorbed to provide high specificity.

#### Secondary antibodies with HRP label

Ig	Species Reactivity	Host	Cross Absorbation	Conjugate	Size	Cat #
IgG (H&L)	Rabbit	Goat	minimum reactivity to bovine, chicken, horse, human, mouse, pig & rat	HRP	1 mg	A120-201P
IgG (H&L)	Mouse	Goat	minimum reactivity to bovine, chicken, horse, human, pig, rabbit & rat	HRP	1 mg	A90-516P
lgG (H&L)	Rat	Goat	minimum reactivity to bovine, chicken, goat, human, mouse, rabbit & sheep	HRP	1 mg	A110-305P

Secondary antibodies with fluorochrome label, suitable for detection with for example the Licor system.

lg	Species Reactivity	Host	Cross Absorbation	Conjugate	Size	Cat #
IgG (H&L)	Rabbit	Goat	minimum reactivity to bovine, chicken, horse, human, mouse, pig & rat	DyLight® 680	0.5 mg	A120-201D6
IgG (H&L)	Rabbit	Goat	minimum reactivity to bovine, chicken, horse, human, mouse, pig & rat	DyLight® 800	1 mg	A120-201D8
IgG (H&L)	Mouse	Goat	minimum reactivity to bovine, chicken, horse, human, pig, rabbit & rat	DyLight® 680	0.5 mg	A90-516D6
IgG (H&L)	Mouse	Goat	minimum reactivity to bovine, chicken, horse, human, pig, rabbit & rat	DyLight® 800	1 mg	A90-516D8

The tables above only present a few recommended examples of secondary antibodies. Numerous of others options with regard to reactivity and host species are available. Search at www.nordicbiosite.com or contact us for help in finding the right secondary antibody.

# **Detection Reagents**

#### Easy-WESTERN

Easy-WESTERN is developed based on MAD (multi-purpose antibody detecting) technology. The main component of MAD is protein particles which have the ability to bind Fc region of antibodies. Each particle has approximately 100 Fc-binding proteins and is labeled with around 50 HRP molecules. Thus Easy-WESTERN provides quick signal enhanced detection of primary antibodies with higher sensitivity without using secondary antibodies.

Easy-WESTERN Super (for high sensitivity detection and signal enhancement)

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181-BCL-EZS01	Easy-WESTERN Super basic	MAD reagent, dilution buffer, manual-S, for 50 tests
181-BCL-EZS02	Easy-WESTERN Super marker detector set	basic + marker detector, for 50 tests
181-BCL-EZS04	Easy-WESTERN Super mouse enhancer set	basic + mouse IgG enhancer, for 50 tests
181-BCL-EZS03	Easy-WESTERN Super full set	basic + marker detector + mouse IgG enhancer, for 50 tests

Easy-WESTERN Quick (for one-step quick detection, multi-antigen simultaneous detection)

181-BCL-EZQ01	Easy-WESTERN Quick basic	MAD reagent, dilution buffer, manual-Q, for 50 tests
181-BCL-EZQ02	Easy-WESTERN Quick marker detector set	basic + marker detector, for 50 tests
181-BCL-EZQ04	Easy-WESTERN Quick mouse enhancer set	basic + mouse IgG enhancer, for 50 tests
181-BCL-EQS03	Easy-WESTERN Quick full set	basic + marker detector + mouse IgG enhancer, for 50 tests

### Chemiluminiscent HRP Substrate/ECL

BioSite stabilized LumiFemto Luminol based substrates can detect horseradish peroxidase at the femtogram level. BioSite chemiluminescent substrates provide superior sensitivity compared to competitor products. These reagents are stable for 30 to 45 days after mixing at 4°C. The substrate is supplied as two components.

LumiFemto Membrane / Blotting Chemiluminescent Peroxidase Substrate (LUMF - Two Components A+B).

Size	Cat #
50 ml	LUMF-50
100 ml	LUMF-100
200 ml	LUMF-200
500 ml	LUMF-500
1,000 ml	LUMF-1000

# Films

- Standard High Contrast, Double Emulsion Blue Film
- High Sensitivity with Low Background
- Optimized for Enhanced Chemiluminescence (ECL)

PerfectFilm, 12,7 x17,8 cm, (100 sheets), cat # B557

PerfectFilm, 20,3 x 25,4 cm, (100 sheets), cat # B581





# Tell us more about your research — we can help you reach your goals



info@nordicbiosite.com www.nordicbiosite.com



