

Product datasheet

Membrane Fraction WB Cocktail ab140365

2 References 4 Images

Overview

Product name	Membrane Fraction WB Cocktail
Sample type	Cell culture extracts, Tissue Extracts, Cell Lysate
Species reactivity	Reacts with: Mouse, Rat, Human
Product overview	ab140365 contains 5 mAbs each targeting proteins located in different compartments of the cell. The presence of plasma membrane is shown by Anti-Sodium Potassium ATPase; endoplasmic reticulum is detected by GRP78; mitochondrial by ATP5A; cytosol by Anti-GAPDH; and nucleus by Anti-Histone H3 (di methyl K9). This cocktail is suitable for detecting the purity of cellular fractions using kit ab139409 .
Tested applications	Suitable for: WB

Properties

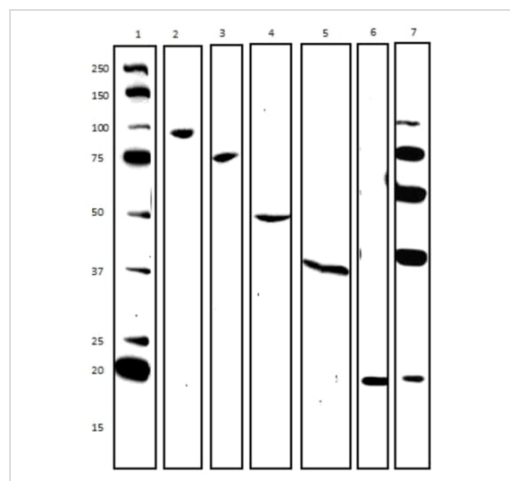
Storage instructions Store at -20°C. Please refer to protocols.

Components	200 µl
2500X HRP Conjugated Secondary Antibody Cocktail	1 x 60µl
250X Membrane Fraction Western Blot Cocktail	1 x 200µl

Applications

The Abpromise guarantee Our [Abpromise guarantee](#) covers the use of ab140365 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Primary antibody cocktail = 1/250 dilution Secondary antibody cocktail = 1/2500 dilution



Membrane Fraction Western Blot Cocktail - Component Separation

Performed under reducing conditions. All blocking and antibody incubation steps were done in 5% milk in TBST. Developed using the ECL technique. Exposure time: 5 mins.

Sample 1: Marker

Samples 2-5: HeLa Cell Lysate – 20 µg

Primary:

Lane 1: none

Lane 2: Anti- Sodium Potassium ATPase antibody – Plasma Membrane Marker

Lane 3: Anti-GRP78 antibody – Endoplasmic Reticulum Marker

Lane 4: Anti- ATP5a antibody – Mitochondrial Membrane Marker

Lane 5: Anti- GAPDH antibody – Cytosolic Marker

Lane 6: Anti- Histone H3 (di methyl K9) antibody – Nuclear Marker

Lane 7: Assembled Membrane Antibody Cocktail

Secondary:HRP conjugated secondary antibody cocktail

Predicted/observed band sizes:

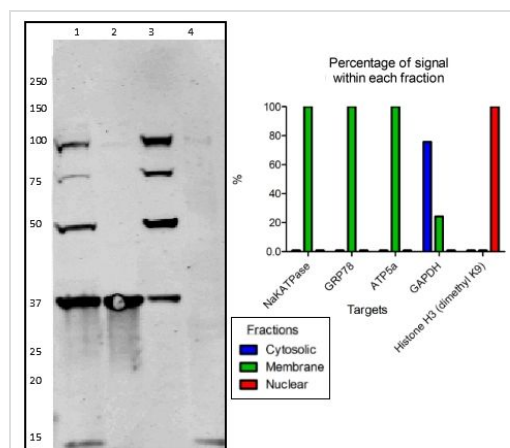
Sodium Potassium ATPase = 112 kDa/100 kDa

GRP78 band size: 78 kDa / 75 kDa

ATP5A band size: 60 kDa / 60 kDa

GAPDH band size: 37 kDa / 38 kDa

Histone H3 (di methyl K9) band size: 17 kDa / 17 kDa



Membrane Fraction Western Blot Cocktail - Specificity

Developed by IR scanning. HeLa cell lysate prepared using Kit [ab139409](#). Exposure time: 5 mins . Samples run on a 4-20% gradient acrylamide gel. All blocking and antibody incubation steps were done in 5% milk, 20mM Tris-HCl, 0.1% TWEEN-20

Lane 1: Marker

Lane 2 : HeLa Whole Cell Lysate

Lane 3 : HeLa Cytosolic Fraction Lysate

Lane 4 : HeLa Membrane Fraction Lysate

Lane 5 : HeLa Nuclear Fraction Lysate

Primary antibody: Membrane Fraction Western Blot cocktail

Secondary: IR800 GAM (1/10000) and IR690 GAR (1/10000)

Predicted/Observed band sizes:

Sodium Potassium ATPase: 112 kDa/100 kDa

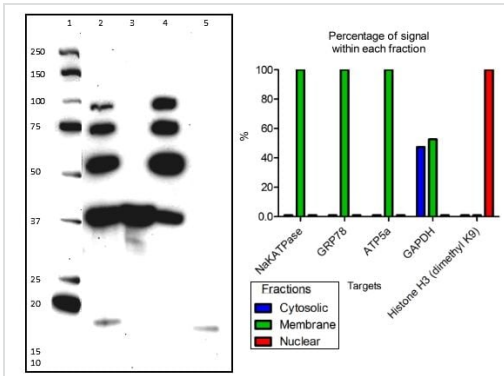
GRP78: 78 kDa/75 kDa

ATP5A: 60 kDa/60 kDa

GAPDH: 37 kDa/37kDa

Histone H3 (di methyl K9): 17 kDa/17kDa

Notes: Percentage of antibody signal (Y axis) represents the signal present in individual fractions as a proportion of the sum of all three fractions (cytoplasmic, membrane and nuclear)



Membrane Fraction Western Blot Cocktail - Specificity

Developed using ECL technique under reducing conditions. HeLa lysate prepared using the Membrane Fractionation Kit ([ab139409](#)).

Exposure time: 5 mins. on a 4-20% gradient acrylamide gel, blocking and antibody incubation steps done in 5% milk, 20mM Tris-HCl, 0.1% TWEEN-20

Lane 1: Marker

Lane 2 : HeLa Whole Cell Lysate

Lane 3 : HeLa Cytosolic Fraction Lysate

Lane 4 : HeLa Membrane Fraction Lysate

Lane 5 : HeLa Nuclear Fraction Lysate

Primary antibody: Membrane Fraction Western Blot cocktail

Secondary: HRP conjugated secondary antibody cocktail

Predicted/Observed band sizes:

Sodium Potassium ATPase: 112 kDa/100 kDa

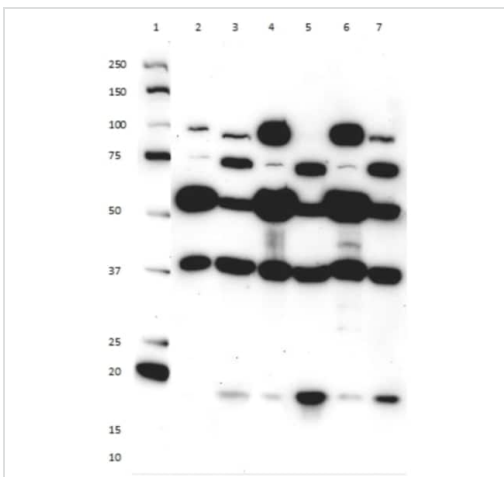
GRP78: 78 kDa/75 kDa

ATP5A: 60 kDa/60 kDa

GAPDH: 37 kDa/37kDa

Histone H3 (di methyl K9): 17 kDa/17kDa

Notes: Percentage of antibody signal represents signal present in individual fractions as a proportion of the sum of all three fractions (cytoplasmic, membrane and nuclear)



Membrane Fraction Western Blot Cocktail - Cross Reactivity

Samples run on a 4-20% gradient acrylamide gel. Performed under reducing conditions. All blocking and antibody incubation steps were done in 5% milk, 20mM Tris-HCl, 0.1% TWEEN-20.

Developed using the ECL technique. Exposure time: 5 mins.

Lane 1: Marker

Lane 2: Human heart homogenate Lysate – 20 µg

Lane 3: HeLa Cell Lysate – 20 µg

Lane 4: Mouse heart homogenate Lysate - 20 µg

Lane 5: NIH3T3 Cell Lysate – 20 µg

Lane 6: Rat heart homogenate Lysate – 20 µg

Lane 7: H9C2 Cell Lysate – 20 µg

Primary antibody: Membrane Fraction Western blot cocktail at 1/250 dilution

Secondary antibody: HRP conjugated secondary antibody cocktail

at 1/2500 dilution

Predicted/observed band sizes:

Sodium Potassium ATPase = 112 kDa/100 kDa

GRP78 band size: 78 kDa / 75 kDa

ATP5A band size: 60 kDa / 60 kDa

GAPDH band size: 37 kDa / 38 kDa

Histone H3 (di methyl K9) band size: 17 kDa / 17 kDa

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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