

# Total OXPHOS Blue Native WB Antibody Cocktail ab110412

★★★★★ [1 Abreviews](#) [46 References](#) [1 Image](#)

### Overview

<b>Product name</b>	Total OXPHOS Blue Native WB Antibody Cocktail
<b>Assay type</b>	Quantitative
<b>Species reactivity</b>	<b>Reacts with:</b> Mouse, Rat, Cow, Human
<b>Product overview</b>	Blue Native polyacrylamide gel electrophoresis (BN-PAGE) is a simple and effective way to subfractionate mitochondrial proteins as intact complexes on a single gel (in one dimension). It can be used to detect altered assembly of these complexes arising from mutations in subunits, mutations in assembly factors, or mtDNA depletion. This type of analysis has been performed with biopsy samples, platelets and fibroblast cells from patients with suspected mitochondrial diseases.

In this method, multisubunit enzymes bind a charged dye Coomassie brilliant blue which allows their electrophoretic separation in the first dimension by the size of the complex. Complexes I-V with masses ranging from 950K to 200K are well resolved in the first dimension. The separated proteins can then be transferred to nitrocellulose membrane/PVDF by electrophoresis and Complexes I, II, III, IV and ATP synthase can be detected by mAbs against C1-NDUFA9 [ab14713](#)(MS111), CII-70 kDa subunit [ab14715](#)(MS204), CIII-Core protein 2 [ab14745](#)(MS304), CIV-subunit IV [ab14744](#)(MS407) and CV-alpha subunit [ab14748](#)(MS507) respectively. Such one dimensional gels, are best analyzed by using single mAbs against each complex. Other Complex I mAbs are available for BN-PAGE, specifically anti-GRIM-19 [ab110240](#)(MS103) and anti-20 kDa [ab110242](#)(MS105). A sample of purified bovine heart mitochondria [ab110338](#)(MS802) is available as a BN-PAGE control sample.

Sometimes a greater separation of enzymes is necessary - it is possible to separate the proteins within each individual complex. To do this, blotting is NOT performed after the first (NATIVE) dimension, instead gels are turned 90 degrees and run in a perpendicular second dimension which is denaturing (NON-NATIVE). In this way the protein subunits within each complex are separated. Abcam provides an optimized pre-mixed cocktail of the mAbs to SIMULTANEOUSLY detect Complexes I-V after 2nd dimension blotting (specifically the cocktail contains [ab14713](#) (MS111), [ab14715](#) (MS204), [ab14745](#) (MS304), [ab14744](#)(MS407) and [ab14748](#) (MS507).

### Cocktail Antibodies:

**Mouse monoclonal [20C11B11B11] to (C-I) NDUFA9 ([ab14713](#)):**

Amount: 120 µg

Working Concentration: 2 µg/ml

**Mouse monoclonal [2E3GC12FB2AE2] to (C-II-70) SDHA ([ab14715](#)):**

Amount: 6 µg

Working Concentration: 0.1 µg/ml

**Mouse monoclonal [13G12AF12BB11] to (C-III-Core 2) UQCRC2 ([ab14745](#)):**

Amount: 60 µg

Working Concentration: 1 µg/ml

**Mouse monoclonal [20E8C12] to (C-IV-subunit IV) COX IV([ab14744](#)):**

Amount: 60 µg

Working Concentration: 1 µg/ml

**Mouse monoclonal [15H4C4 ] to (C-V-alpha) ATP5A ([ab14748](#)):**

Amount: 60 µg

Working Concentration: 1 µg/ml

**Notes****Related products**

Review the [mitochondrial assay guide](#), or the full [metabolism assay guide](#) to learn about more assays for metabolites, metabolic enzymes, mitochondrial function, and oxidative stress, and also how to assay metabolic function in live cells using your plate reader.

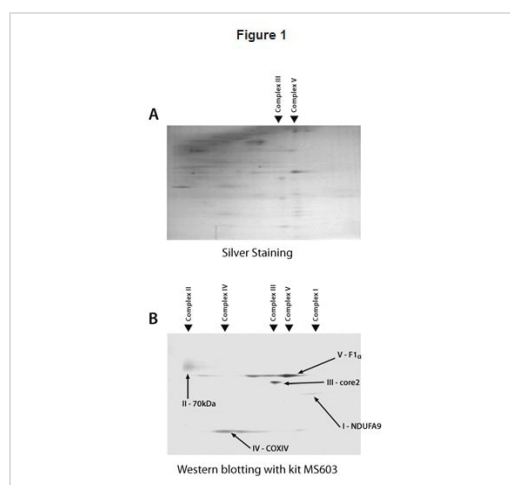
**Tested applications****Suitable for:** WB**Properties****Storage instructions** Store at -80°C. Please refer to protocols.

Components	300 µg
Antibody Cocktail	1 x 300µg
Bovine heart mitochondria control	1 unit

**Applications****The Abpromise guarantee** Our [Abpromise guarantee](#) covers the use of ab110412 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
<b>WB</b>		Use a concentration of 6 µg/ml. The antibody cocktail (1.5 mg/mL) should be diluted 250x to a final working concentration of 6.0 µg/mL for Western blotting



Western blot - MitoProfile® Total OXPHOS Blue  
Native WB Antibody Cocktail (ab110412)

Two dimension Blue Native PAGE analysis of fibroblasts that are (A) normal and (B) complex I deficient. It is clear that the complex I deficient cell line shown in B shows no detectable level of complex I. However, all other OXPHOS complexes appear normal including a minor amount of heterocomplex formed between complexes III and IV which is a well documented species. Only a minor mitochondrial enrichment was performed upon these samples cell lines. Each sample represents only 8% of confluent cells taken from a 10 cm diameter tissue culture dish.

MWs:

(C-I) NDUFA9 - 36kDa

(C-II-70) SDHA - 70kDa

(C-III-Core 2) UQRC2) - 45kDa

(C-IV subunit IV) COX IV - 15kDa

(C-V alpha) ATP5A - 55kDa

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

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