MitoSciences

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TECHNICAL DATA SHEET

MetaPath™ MitoDisease 4-Plex Dipstick Array

Catalog # MSX31

Rev.3

LOT #:

APPLICATIONS:

Quantitative analysis of Complex I, Complex IV, Frataxin and PDH. For measuring (i) the inhibition of mitochondrial protein translation in mitochondrial genetic disease, (ii) assembly of PDH complex a common mitochondrial deficiency and (iii) expression of frataxin the cause of Friedreich's ataxia.

SPECIES CROSS-REACTIVITY:

Human

KIT COMPONENTS:

Item	MSX31-30	MSX31-90
Dipsticks	30	90
Gold-conjugated antibody (dried in microplate wells)	30 wells	90 wells
Buffer A (Extraction buffer)	15 mL	45 mL
Buffer B (Blocking solution)	1 mL	3 mL
Buffer C (Wash buffer)	2 x (1 mL)	6 x (1 mL)

STORAGE CONDITIONS:

Store dipsticks and gold-conjugated antibody in their provided storage containers at room temperature, out of direct sunlight.
Store Buffers A, B, and C at 4°C or at -20°C for long term storage.

COUNTRY OF ORIGIN:

USA

BACKGROUND:

The MetaPath™ MitoDisease Dipstick Array (MSX31) is a rapid and simple test for determining four mitochondrial protein parameters: OXPHOS Complex I, OXPHOS Complex IV, pyruvate dehydrogenase, and frataxin. The kit determines whether mitochondrial biogenesis has been up-regulated or down-regulated in human samples as a result of drug interactions or conditions such as oxidative stress and mitochondrial genetic disease.

Each of the dipsticks in the kit contains an anti-Complex I mAb and an anti-Complex IV mAb for monitoring specific defects in either complex, defects in mtDNA or dysfunctional protein synthesis by mitochondrial ribosomes. Each dipstick also contains an anti-PDH mAb and an anti-frataxin mAb these proteins can be used as mitochondrial reference proteins to compare against Complexes I and IV, PDH and frataxin are also affected in the specific mitochondrial diseases pyruvate dehydrogenase complex disease (PDCD) and Friedreich's ataxia (FA), respectively.

The assay works with very small amounts of sample material, including cheek swabs or single drops of blood, and results can be generated in under 30 minutes. The assay also works on cultured cells and is suitable for any situation in which speed and simplicity are required.

The method is based on an immunological sandwich assay (see Figure 1). Each enzyme is bound by two unique monoclonal antibodies. To accomplish this, one antibody is gold labeled and mixed with the sample. The other is embedded in the strip in one of four vertically separated zones. As the sample wicks past each zone the

corresponding enzyme (now gold-labeled) is trapped, this results in a red colored band which is directly proportional in intensity to the amount of each enzyme captured.

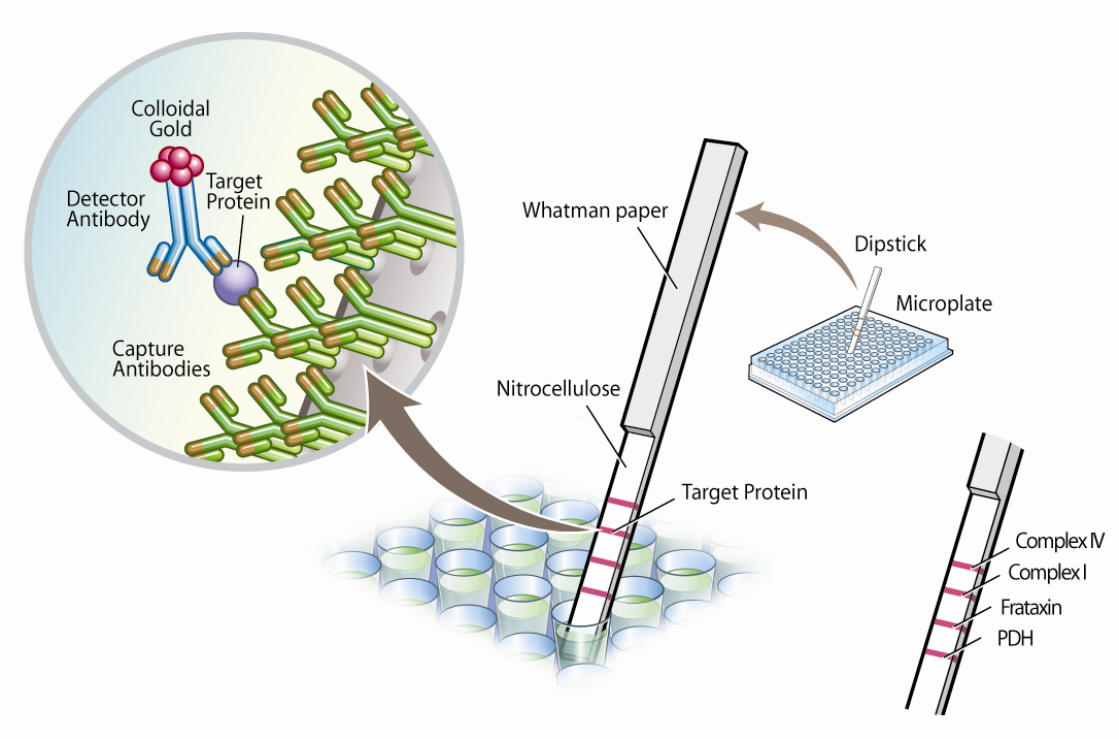

Figure 1. Assay principle.

Standard curves of Complex I, IV, PDH and frataxin levels are generated by the user with a control dilution series of normal sample material. The signal intensities of the four bands on the dipsticks are measured by a dipstick reader (MitoSciences' MS1000 is recommended) or may be analyzed by another imaging system. The levels of all 4 enzymes in an experimental sample are then measured by interpolating their signal intensities to the standard curves. The levels and ratios of the enzymes in experimental samples are then compared with the control dilution series.