abcam

Product datasheet

Pyruvate dehydrogenase (PDH) Enzyme Activity Dipstick Assay Kit ab109882

27 References 3 图像

概述

产品概述

产品名称 Pyruvate dehydrogenase (PDH) Enzyme Activity Dipstick Assay试剂盒

样品类型 Cell culture extracts, Tissue 检测类型 Sandwich (quantitative)

种属反应性 与反应: Mouse, Rat, Cow, Human, Pig

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ab109882 is used to quantify the activity of the PDH enzyme complex from human, bovine, mouse, rat and pig samples. This is accomplished by immunocapturing PDH with an anti-PDH antibody immobilized on a defined section of a dipstick. The enzyme complex is immunocaptured in its native form and activity is measured by the reaction scheme. PDH activity is visualized by coupling PDH-dependent production of NADH to the reduction of NBT in the presence of excess diaphorase, forming an insoluble intensely colored precipitate at the capture line. The signal intensity is measured by a dipstick reader or analyzed by other imaging systems such as a flatbed scanner.

Purified mitochondria can be used in this assay. However, homogenized tissue and whole cells can also be used without the need for mitochondrial isolation.

Pyruvate dehydrogenase activity is regulated by PDH kinase and PDH phosphatase. Cells grown in glucose media derive most of their energy from glycolysis; therefore, most of their PDH complex may be in an inactive phosphorylated form at the time of isolation. Growing cells in alternate carbon sources such as glutamine/galactose may up-regulate PDH activity. This kit does not include PDH kinases, PDH phosphatases or their respective inhibitors. These may be incorporated into the assay at the user's discretion.

Store dipsticks at room temperature out of direct sunlight in their provided desiccant container. Avoid high humidity conditions.

Store 20X Reagent Mix, Detergent, and Diaphorase at -80°C, but these may be aliquoted upon receipt to prevent freeze/thaw cycles.

Store NBT in the dark at -80°C.

经测试应用 适用于: Functional Studies

平台 Reagents

说明

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存放说明

Please refer to protocols.

组件	30 tests
20X Reagent Mix	1 x 600µl
96-well microplate	2 units
Blocking solution	1 x 2ml
Detergent	1 x 1ml
Diaphorase	1 x 400µl
Dipsticks	1 x 30 tests
NBT	1 x 40µl
Sample buffer	1 x 30ml

应用

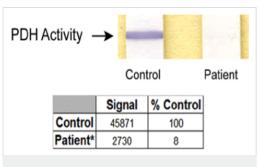
The Abpromise guarantee

Abpromise™承诺保证使用ab109882于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Functional Studies		Use at an assay dependent dilution.

图片



Pyruvate dehydrogenase (PDH) Enzyme Activity Dipstick Assay Kit (ab109882) Figure 2. An example using the ab109882 to measure PDH activity in human fibroblast sample. Based on the standard curve, 160 μg of protein extract were loaded onto a dipstick for each sample. The figure shows two developed dipsticks, a control sample and a sample from a patient with a mutation in the E1-alpha subunit of PDH. The analysis of the signal intensity and interpolation from the standard curve showed that the patient sample had 8% the PDH activity of the control.

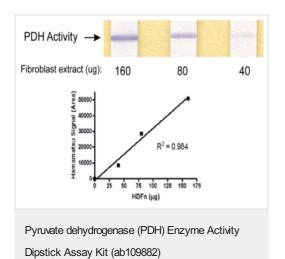
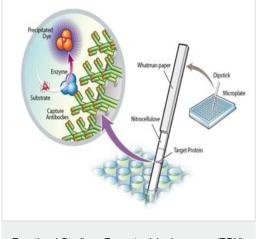


Figure 1. An example using the ab109882 to measure PDH activity in human fibroblast sample. Developed dipsticks from a 1:2 dilution series using a positive control sample and the associated standard curve. Starting material was 160 μg of fibroblast protein extract in a 100 μL reaction volume (50 μL sample in Sample buffer plus 50 μL of Blocking solution). The dipsticks were developed for 1 hour in Activity buffer.



Functional Studies - Pyruvate dehydrogenase (PDH) Enzyme Activity Dipstick Assay Kit (ab109882) Abcam's enzyme activity assays apply a novel approach, whereby target enzymes are first immunocaptured from tissue or cell samples before subsequent functional analysis. Dipstick ELISA Kits extend this concept by utilizing the well-established lateral flow concept, wherein capture antibodies are striped onto nitrocellulose membrane and a wicking pad draws the sample through the antibody bands. All of our ELISA kits utilize highly validated monoclonal antibodies and proprietary buffers, which are able to capture even very large enzyme complexes in their fully-intact, functionally-active states.

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

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