CREATIVE BIOMART®
Assay Kit

NOS Activity Assay Kit

Product Information

Cat.No.

Kit-0631

Product Overview

The NOS activity assay kit measures NOS activity by monitoring the conversion of radiolabeled arginine to citrulline. This assay is simple, sensitive, and specific for NOS activity and can be used with both crude and purified enzyme preparations. The kit includes sufficient materials and reagents for 50 total reactions. Radiolabeled arginine and NADPH are not included with the kit.For routine assays, radioactive arginine is added to protein extracts or purified NOS samples. After incubation, the reactions are stopped with a buffer containing ethylenediaminetetraacetic acid (EDTA), which chelates the calcium required by nNOS (neuronal NOS; NOS I) and eNOS (endothelial NOS; NOS III) and, consequently, inactivates the enzyme. In the case of iNOS (inducible NOS; NOS II), the low pH of the Stop Buffer, pH 5.5, stops the enzyme-catalyzed reaction. Equilibrated Resin, which binds to the arginine, is added to the sample reactions and the reactions are then pipetted into spin cups. The citrulline, being ionically neutral at pH 5.5, flows through the cups completely. NOS activity is then quantitated by counting the radioactivity in the eluate.

Usage

Please read these instructions carefully before beginning this assay.For research use only. Not for human or diagnostic use.

Storage

Stability: 1 year;Storage: -80°C;This kit will perform as specified if stored as listed in the Materials Supplied and used before the expiration date indicated on the outside of the box.

Kit Components

Kit will arrive packaged as a -80°C kit. For best results, remove components and store as stated below.iNOS Positive Control: 50 μL, -80°C;Calmodulin (1 μM): 200 μL, -20°C;Reaction Buffer (2X): 1.25 ml, -20°C;L-NNA (10 mM): 100 μL, -20°C;Homogenization Buffer (10X): 50 ml, Room Temperature;Stop Buffer: 25 ml, Room Temperature;Equilibrated Resin: 5 ml, Room

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Temperature;Calcium Chloride (6 mM): 400 μL, Room Temperature;Spin cups and cup holders 50 Room Temperature

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