

ReadiUse™ NADP Regenerating Kit

Catalog number: 15266

Unit size: 200 Tests

Component	Storage	Amount
Component A: NADP Regenerating Buffer I	Freeze (<-15 °C), Minimize light exposure	10 mL
Component B: NADP Regenerating Buffer II	Freeze (<-15 °C), Minimize light exposure	10 mL
Component C: NADP Regenerating Enzyme Mix	Freeze (<-15 °C), Minimize light exposure	1 vial
Component D: NADPH	Freeze (<-15 °C), Minimize light exposure	2 vials

OVERVIEW

NADP is the electron acceptor for biosynthetic reactions and dehydrogenation reactions. NADP is used for anabolic pathways, such as lipid synthesis, cholesterol synthesis and fatty acid chain elongation. It is also a necessary cofactor in many xenobiotic metabolism reactions. In chloroplasts, NADP is reduced to NADPH by ferredoxin-NADP reductase in the last step of the electron chain in photosynthesis reactions. Many oxidoreductases and all ligases use NADP/NADPH as coenzyme. NADP can also be used in the determination of amylase, creatine kinase, glucose-6-phosphate dehydrogenase, isocitrate dehydrogenase. AAT Bioquest's ReadiUse™ NADP Regenerating Kit provides a flexible prepackaged NADP regenerating system to accommodate various experimental designs. It uses two ready-to-use solutions to regenerate NADP by a simple mixing. This kit can be used for all NADP-requiring systems (such as: 12-Ketocholestenoid synthesis, Ferredoxin Reductase System). 300-500 enzyme assays can be performed using this kit.

AT A GLANCE

Important Thaw all the components at room temperature before use.

PREPARATION OF STOCK SOLUTIONS

Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles.

1. NADPH solution (40X):

Add 0.5 mL H₂O into one vial of NADPH (Component D) to make 40X NADPH solution.

Note One vial of NADPH is for making 10 mL NADP Regenerating Solution.

2. NADP Regenerating Enzyme Mix solution (400X):

Add 100 µL H₂O into the vial of NADP Regenerating Enzyme Mix (Component C) to make 400X NADP Regenerating Enzyme Mix solution. Protect from light.

PREPARATION OF WORKING SOLUTION

Add 5 mL of NADP Regenerating Buffer I (Component A), 5 mL of NADP Regenerating Buffer II (Component B), 0.5 mL of 40X NADPH solution, and 50 µL of 400X NADP Regenerating Enzyme Mix solution to make a total volume of 10.55 mL 2X NADP regenerating solution. Keep from light.

Note Activated NADP regenerating solution is not stable, and should be prepared freshly before use. 10 mL of NADP Regenerating Solution is enough for 1 plate.

SAMPLE EXPERIMENTAL PROTOCOL

1. Add equal volume of 2X NADP Regenerating solution into the desired assay system.

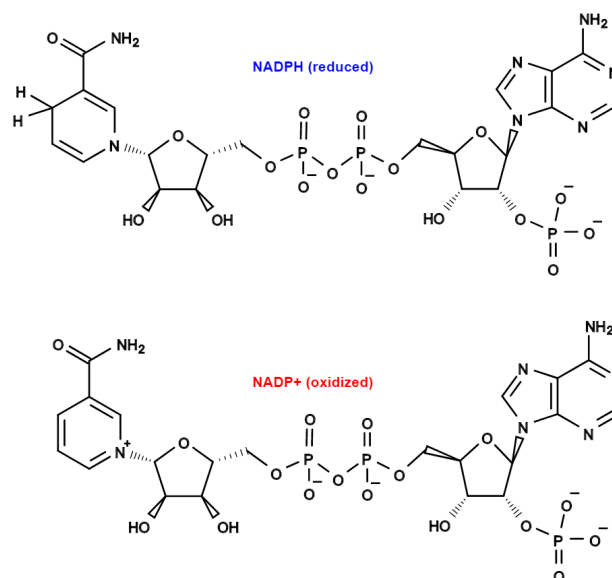
EXAMPLE DATA ANALYSIS AND FIGURES


Figure 1. Chemical structure for ReadiUse™ NADP Regenerating Kit

DISCLAIMER

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