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Product Information

Nitrate Reductase (NAD[P]H)
from *Aspergillus niger*

Catalog Number **N7265**
Storage Temperature –20 °C

CAS RN 9029-27-0
EC 1.7.1.2
Synonym: NAD(P)H:Nitrate oxidoreductase

Product Description

The molecular mass of this enzyme is 180 kDa (gel filtration) with two subunits of 59 kDa and 38 kDa (SDS-PAGE).[1] Another reference noted only one subunit of 54 kDa (SDS-PAGE).[2] This product showed a major band at 56.9 kDa and minor band at 32.5 kDa (SDS-PAGE).

Isoelectric point: 6.12±0.05 (22 °C for the *Aspergillus nidulans* enzyme)[2]

Nitrate reductase from *Aspergillus* has a K_M for nitrate of ~199 µM in phosphate buffer, pH 7.5, containing 100 µM NADPH, and 2.5 µM FAD at 25 °C.[3]

Nitrate reductase has been used in a spectrophotometric assay for nitrate[3] and a fluorometric assay for the measurement of nitrite in biological samples, detecting 10 nM nitrite (50–100 fold more sensitive than the Griess assay).[4]

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

Store the product at –20 °C.

When the enzyme was dissolved in distilled water for 30 minutes at 23 °C, then diluted to 3.5 units/ml in 0.14 M potassium phosphate, pH 7.5, and stored on ice, it remained active for at least 12 hours.[3] Another reference indicated that if dissolved in 150 mM phosphate buffer with 20 µM FAD and 1 mM EDTA, aliquots of the purified enzyme could be stored for several months at –80 °C with only slight loss of activity.[4] Stock solutions of the enzyme at 10 units/ml in water were frozen in 100 µl aliquots at –70 °C with no noticeable change in activity over several months.[5]

References

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2. Steiner, F.X., and Downey, R.J., Isoelectric focusing and two-dimensional analysis of purified nitrate reductase from *Aspergillus nidulans*. Biochim. Biophys. Acta, **706(2)**, 203-211 (1982).
3. Gilliam, M.B. et al., A spectrophotometric assay for nitrate using NADPH oxidation by *Aspergillus* nitrate reductase. Anal. Biochem., **212(2)**, 359-365 (1993).
4. Misko, T.P. et al., A flurometric assay for the measurement of nitrite in biological samples. Anal. Biochem., **214(1)**, 11-16 (1993).
5. Grisham, M.B., et al., Quantitation of nitrate and nitrite in extracellular fluids. Methods in Enzymology, **268**, 237 (1996).

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