BIOCATALYSTS | A BRAIN Group company

Phenylalanine Dehydrogenase

P098P

Features/Benefits

- Neonatal screening enzyme
- Phenylketonuria screening (Pku) test
- Lower false positives than other tests
- Lower total costs than Guthrie method
- Simple and rapid

Phenylketonuria (PKU) is an inborn metabolic disorder where those affected lack the ability to metabolise phenylalanine. If undiagnosed brain and developmental disorders arise, if diagnosed, treated early and subsequently managed then normal development can ensue. Screening of newborns for PKU has been practised in the Western world for some time.

The trend is for growing screening programs worldwide.

The traditional method is the Guthrie microbial test, but this is being replaced by a cheaper, more rapid and sensitive enzyme test based on phenylalanine dehydrogenase which has proved sensitive, reliable and rapid compared to other methodologies. Low cross reactivity results in lower false positives than the Guthrie and fluorometric methods.

Specification

Minimum Activity	6 units of phenylalanine dehydrogenase per mg solid
Specific Activity	> 20 units/mg protein
Unit Definition	1 unit converts 1 µM of NAD+ in the presence of phenylalanine at pH 10.5 and 30°C
Biological Source	*Sporosarcina sp.*
Form	Lyophilised off-white powder
K_M	5 mM (on NAD)
IUB No & Type	11.4.1.20 / L-phenylalanine: NAD oxido-reductase
Optimum pH Range	10.0 - 11.0
Optimum Temperature Range	35 - 40°C

Phenylalanine dehydrogenase (PheDH) is an enzyme suitable for use in neonatal screening kits. This enzyme converts phenylalanine to phenylpyruvate in the presence of NAD+ which acts as an electron acceptor. Electrons from the reduced NAD+ i.e., NADH are transferred to a substrate such as Iodonitrotetrazolium hydrochloride (INT) via the action of the enzyme diaphorase. This process results in the formation of the red formazan dye which can be quantified spectrophotometrically. Blood samples are taken from neonatals via a “heel prick”, and the blood sampled dabbed onto a filter paper. Where this enzyme is used in a commercial kit the method utilises a blood spot on a filter paper which has been dipped in methanol to fix blood proteins. Amino acids are eluted from the blood spot into water. A sample of the eluate is incubated with PheDH, NAD+, Diaphorase and INT with the result determined by spectrophotometric measurement. This might use an instrument such as the centrifugal analyser or a microplate reader. The result will be obtained by reference to a set of standards. The disorder will be indicated by above normal levels of the phenylalanine. The exact levels of enzyme, buffering reagents and conditions to be used in such a kit to obtain the desired measuring range and reliability will have to be determined in studies. The temperature and pH optimum indicated for this product are conditions where the product gives maximum activity as measured using the Biocatalysts’ assay procedure. Further information on PKU can be found in the references cited below.

Dooley K.C., 1992, Clin. Biochem., 25, pp 271 - 275. Campbell R.S., Hollifield R.D., Varsani H., Milligan T.P., Brearley G. and Price C.P., 1994, Anal. Clin. Biochem., 31, pp 140-146. Campbell R.S., Brearley G., Varsani H., Morris H.C., Milligan T.P., Halls K. et. al., Clin. Chimica. Acta., 1992, 210, pp197 - 210. Wendel U., Hummel W. and Langenbeck U., 1989, Anal. Biochem., 180, pp 91- 94.

Health & Safety

Always read the Material Safety Datasheet (MSDS) before use and retain. If you are in any doubt about recommended product handling and safety, please contact Biocatalysts before use. Generally, when using enzymes avoid contact with the skin and eyes and do not breathe dusts or aerosols containing them.

GM Status

This product does not contain GMMs or genetically modified material.

Food Status

This product has not been produced as a food grade product but has been manufactured under ISO 9001 accreditation.

Quality

1. Good Manufacturing Practice (GMP) - The Company’s integrated management system encompasses Total Quality, Health and Safety, Food Safety and GMP.
2. Biocatalysts Ltd is certified to ISO9001, ISO14001, ISO45001 and FSSC 22000.

Availability

Available in 1,000- and 10,000-unit packs.

Storage

This product is fully active when stored desiccated at -20°C.

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