

6 Henshaw Street

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Human FMO1 SUPERSOMES™

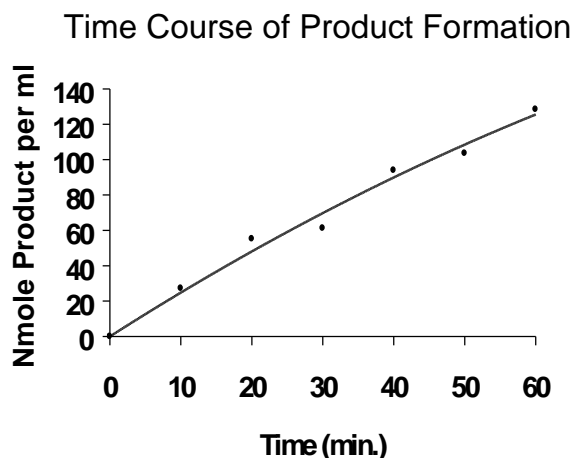
Catalog Number456241
Lot Number2408080

Storage Conditions..STORE AT -80°C
Date Released2024 August
Expiration Date2034 August

Package Contents.....2.5 mg protein in 0.5 ml
Protein Content.....5.0 mg/ml in 100mM potassium phosphate (pH 7.4)
Methyl p-Tolyl Sulfide Oxidase Activity ...5000 pmol product/(min x mg protein)
FAD Content¹380 pmol FAD/mg protein

This activity is catalyzed by FMO1 which is expressed from human FMO1 cDNA using a baculovirus expression system. Baculovirus infected insect cells (BTI-TN-5B1-4) were used to prepare these microsomes. A microsome preparation using wild type virus (Catalog No. 456201) should be used as a control for native activities.

METHOD: A 0.25 ml reaction mixture containing 50 µg protein, 0.065 mM NADP⁺, 3.3 mM glucose-6-phosphate, 0.4 U/ml glucose-6-phosphate dehydrogenase, 3.3 mM magnesium chloride, and 2.0 mM methyl p-tolyl sulfide in 0.05 M glycine (pH 9.5) was incubated at 37°C for 10 min. After incubation, the reaction was stopped by the addition of 75 µl acetonitrile and centrifuged (10,000 x g) for 5 minutes. 100 µl of the supernatant was injected into a 4.6 x 250 mm 5µ C18 HPLC column and separated at 45°C with a mobile phase initially of 46% methanol increasing to 55% methanol over 7 min. (the substrate was then eluted with 100% methanol) and at a flow rate of 1.0 ml per min. The product was detected by its absorbance at 237 nm and quantitated by comparing the absorbance to a standard curve of methyl p-tolyl sulfoxide.



ADVICE

- Thaw rapidly in a 37°C water bath. Keep on ice until use.
- Aliquot to minimize freeze-thawing cycles. Less than 20% of the catalytic activity is lost after 6 freeze thaw cycles.
- Metabolite production is linear with respect to enzyme concentration up to at least 0.6 mg per ml. Metabolite production with methyl p-tolyl sulfide is approximately linear for 40 minutes (see graph above). Other substrates may not exhibit similar linearity with respect to incubation time.

¹ FAD content was determined by a method based on the one described by Lang et al. in Biochemical Pharmacology **56**, 1005 (1998).

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INSECT CELL MICROSOMES

HAZARD WARNING:

The product was produced using baculovirus (*Autographa californica*) infected insect cells (BTI-TN-5B1-4). This virus is not known to be pathogenic to humans or other mammals.

SAFETY INFORMATION:

Safety assessment indicates this product is not hazardous, therefore no SDS (Safety Data Sheet) is provided. Use standard laboratory practices for the handling and disposal of Biosafety Level 1 materials.

Alexa Silies

Quality Assurance

08/27/2024

Date

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