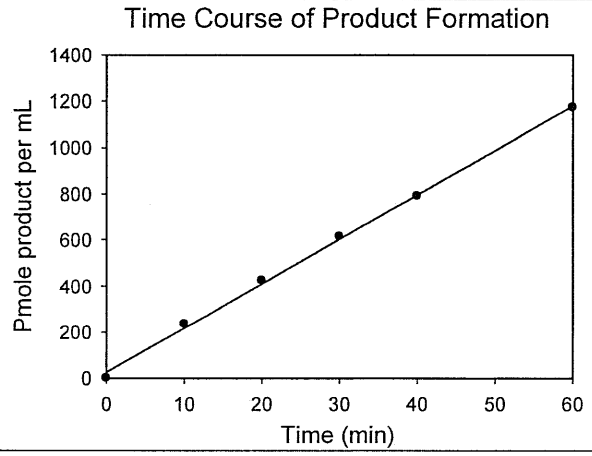
Discovery Life Sciences
6 Henshaw Street
Woburn, MA 01801
Tel: (866) 838-2798
info@dls.com
https://www.dls.com/

Supersomes™ Human UGT2B10

Catalog Number	453323
Quantity / Package	0.5 mL
Protein Content	5.0 mg/mL in 0.1 M Tris (pH 7.5)
Storage Conditions	NA
Storage Temperature	-80°C
Historical Data	

Additional Information

- The amitriptyline N-glucuronidation activity is catalyzed by UGT2B10 which is expressed from human UGT2B10 cDNA using a baculovirus expression system. Baculovirus infected insect cells (BTI-TN-5B1-4) were used to prepare these membranes. A membrane preparation using wild-type virus (**Cat. No. 456400**) should be used as a control for native activities.
- METHOD (amitriptyline N-glucuronidation activity): A 0.2 mL reaction mixture containing 1.0 mg/mL protein, 1 mM uridine diphosphoglucuronic acid (UDPGA), 10 mM magnesium chloride, 0.025 mg/mL alamethicin and 30 µM amitriptyline in 50 mM Tris (pH 7.5) was incubated at 37°C for 60 minutes. After incubation, the reaction was stopped by the addition of 50 µL 94% acetonitrile/6% glacial acetic acid and centrifuged (14,000 × *g*) for 3 minutes. 100 µL of the supernatant was injected into a 4.6 x 250 mm 5µ C18 HPLC column and separated at 45°C using a linear gradient. Initial HPLC conditions were 70% of a 0.1% trifluoroacetic acid/H_2O solution (mobile phase A), and 30% of a solution consisting of 0.1% trifluoroacetic acid in 100% acetonitrile (mobile phase B). The concentration of mobile phase B was increased to 51% over 14 minutes. The HPLC flow rate was 1 mL per minute. The product was detected by its absorbance at 240 nm, and quantitated by comparing to the absorbance of an external standard curve consisting of amitriptyline (parent compound). Previous studies [Breyer-Pfaff U, et al. *Xenobiotica*, 20:727-738 (1990)], and studies at Corning Life Sciences, suggest that the absorption characteristics of aliphatic N+-glucuronides are similar to the aglycone.
- Metabolite production using amitriptyline as a substrate is linear with respect to enzyme concentration up to 1.0 mg/mL (highest concentration tested).
- Metabolite production with amitriptyline is linear for at least 60 minutes (see graph). Other substrates may not exhibit similar linearity with respect to incubation time.

Discovery Life Sciences
6 Henshaw Street
Woburn, MA 01801
Tel: (866) 838-2798
info@dls.com
https://www.dls.com/

Supersomes™ Human UGT2B10

Catalog Number	453323

Safety Information

- Refer to MSDS for safety information.

Recommendation

- Aliquot to minimize freeze-thawing cycles. Less than 5% of the catalytic activity is lost after 8 freeze thaw cycles.
- Thaw rapidly in a 37°C water bath. Keep on ice until use.

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.

Discovery Life Sciences
6 Henshaw Street
Woburn, MA 01801
Tel: (866) 838-2798
info@dls.com
https://www.dls.com/

Supersomes™ Human UGT2B10

Catalog Number	453323
Lot Number	2309201
Manufacturing Date (MONTH/DAY/YEAR)	August/29/2023
Expiration Date (MONTH/DAY/YEAR)	August/29/2033

Assay Results

Enzyme Measured	Assay	Enzyme Activity	Unit
UGT2B10	Amitriptyline N-Glucuronidation	6.3	pmol/(mg × min)

Quality Assurance: _____ Date: 9 October 2023