

3050 Spruce Street, St. Louis, MO 63103 USA
Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757
email: techservice@sial.com sigma-aldrich.com

Product Information

β-Glucuronidase from limpets (*Patella vulgata*)

Catalog Number **G2174** Storage Temperature 2–8 °C

CAS RN 9001-45-0 EC 3.2.1.31

Synonym: β-D-Glucuronide glucuronosohydrolase

Product Description

Glucuronidation, conjugation with glucuronic acid, by the human UDP-glucuronosyltransferase (UGT) family of enzymes plays an important role in the metabolic fate of many drugs and other xenobiotics. This biosynthetic reaction also has a role in the conjugation and excretion of endogenous substrates, such as steroids, bilirubin, and bile acids. UGT activity results in the conjugation of glucuronic acid to substrates containing sulfhydryl, hydroxyl, aromatic amino, or carboxylic acid moieties. The glucuronides formed are more polar (water soluble) than the parent organic substrate and are generally excreted through the kidney.

β-glucuronidase catalyzes the reaction:

β-D-glucuronoside + H₂O → an alcohol + D-glucuronate

β-Glucuronidase from limpets has been shown to be a superior enzyme for the hydrolysis of drug-glucuronides from urine. ^{2,3} This enzyme preparation was found to be cost-effective and thermostable; it could be used at a temperature high enough to allow for a shorter incubation time as compared to the enzyme from snail or bovine. ² Although the exact amount needed will depend on the specific conditions used and must be determined empirically, complete hydrolysis of morphine glucuronide was reported following a 3 hour incubation at 65 °C with 5,000 units of the enzyme per ml of urine. ² Another report found the optimal conditions for hydrolysis of conjugated steroid metabolites to be a one hour incubation at 55 °C at pH 5.2 using 600 units of enzyme per ml of urine. ⁴

 β -Glucuronidase from limpets is a crude solution of enzymes. Many β -glucuronidase preparations may also contain sulfatase activity. For this reason, sulfatase activity is also determined. This product is supplied as an aqueous solution in 0.9% NaCl with 0.02% sodium azide as preservative.

Optimal pH:

glucuronidase activity 4.5–5.0 sulfatase activity ~6.2

<u>Inhibitors</u>: D-glucuronic acid

(Catalog No. G5269) D-galacturonic acid D-glucaro-1,4-lactone

Substrates:

5-Bromo-6-chloro-3-indolyl β-D-glucuronide B4532

6-Bromo-2-naphthyl β-D-glucuronide

5-Bromo-4-chloro-3-indolyl β-D-glucuronide

sodium salt tablet B8174

8-Hydroxyquinoline glucuronide

4-Methylumbelliferyl β -D-glucuronide M9130

4-Nitrophenyl β-D-glucuronide 73677

Glucuronidase Activity: ≥85,000 units per ml

Unit Definition: One Sigma or modified Fishman unit will liberate 1.0 μ g of phenolphthalein from phenolphthalein glucuronide per hour at 37 °C at pH 3.8 (30 minute assay).

Sulfatase Activity: reported on the certificate of analysis

Unit Definition: One unit of sulfatase will hydrolyze 1.0 μ mole of *p*-nitrocatechol sulfate per hour at pH 5.0 at 37 °C.

Precautions and Disclaimer

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

Storage/Stability

Store the product at 2–8 $^{\circ}$ C. When stored at 2–8 $^{\circ}$ C, the enzyme retains activity for at least 3 years.

References

- Tephly, T.R. et al., Adv. Pharmacol., 42, 343-346 (1998).
- 2. Combie, J. et al., Clin. Chem., 28, 83-86 (1982).
- 3. Combie, J. *et al.*, Res. Commun. Chem. Pathol. Pharmacol., **35**, 27-41 (1982).
- 4. Ferchaud, V., *et al.*, Analyst, **125**, 2255-2259 (2000).

RBG,MAM 10/13-1