Product Information

β-Glucuronidase from *Escherichia coli*

Type IX-A, lyophilized powder, 1,000,000-5,000,000 units/g protein

G7396

Product Description

CAS Registry Number: 9001-45-0

Enzyme Commission (EC) Number: 3.2.1.31

Synonyms: β-D-Glucuronide glucuronosohydrolase

Molecular Weight: ~290 kDa (tetramer),[1] 68,259 Da (monomer)[2]

Glucuronidation, or 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.[3] UGT activity results in the conjugation of glucuronic acid to substrates that contain sulfhydryl, hydroxyl, aromatic amino, or carboxylic acid moieties. The resulting glucuronides are more polar (water-soluble) than the parent organic substrate and are generally excreted through the kidney.

β-glucuronidase catalyzes the general reaction:

β-D-glucuronoside + H_2O ↔
an alcohol + D-glucuronate

β-Glucuronidase from *E. coli* is used for the enzymatic hydrolysis of β-glucuronides in urine and other fluids. It does not hydrolyze α-glucuronides or β-glucosides.[4] β-Glucuronidase from *E. coli* has a high rate of hydrolytic activity, and retains this activity during hydrolysis better than similar enzymes that are more sensitive to changes in the concentration of β-glucuronide conjugates. β-Glucuronidase from *E. coli* has been shown to be useful for determining the presence of androsterone, 17-hydroxycorticosteroids, and estriol in urine.[5]

The optimal conditions for the enzymatic hydrolysis of α-hydroxytriazolam, a major triazolam metabolite in human urine, were determined using β-glucuronidase Type IX-A. It was found that a 90-minute incubation of 1 mL of urine with 100 units of the enzyme at 37 °C and pH 5.5-7.8, effectively hydrolyzed the α-hydroxytriazolam given at the clinical dose.[6]

Several references[7-12] have cited use of product G7396 in their protocols.

Precautions and Disclaimer

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.

Product

β-Glucuronidase Type IX-A from *E. coli* is supplied as a powder lyophilized from 10 mM potassium phosphate, 1 mM ethylenediaminetetraacetic acid, and 1 mM dithiothreitol. Polyethylene glycol is added as a stabilizer.

Unlike the enzyme preparation from snail (*Helix pomatia*) that naturally contains β-glucuronidase and sulfatase activities in almost equal amounts, the preparation of β-glucuronidase from *E. coli* is essentially free of sulfatase activity.

Glucuronidase Activity:
1,000,000 - 5,000,000 units/g protein

Unit Definition: One Sigma or modified Fishman unit will liberate 1.0 µg of phenolphthalein from phenolphthalein glucuronide per hour at 37 °C at pH 5.0 (30-minute assay).

Optimal pH: 6-7

Substrates

- 5-Bromo-6-chloro-3-indolyl β-D-glucuronide (Cat. No. B4532)
- 6-Bromo-2-naphthyl β-D-glucuronide (Cat. No. B7877)
- 5-Bromo-4-chloro-3-indolyl β-D-glucuronide sodium salt tablet (Cat. No. B8174)
- 8-Hydroxyquinoline glucuronide sodium salt (Cat. No. 38153)
- 4-Methylumbelliferyl β-D-glucuronide (Cat. No. M9130)
- 4-Nitrophenyl β-D-glucuronide (Cat. Nos. N1627, 73677)

Inhibitors

- D-glucuronic acid (Cat. No. G5269)
- D-galacturonic acid (Cat. No. 48280)
- D-glucaro-1,4-lactone

Solubility

When reconstituted to 5 mg/mL in 75 mM phosphate buffer (pH 6.8), a clear to slightly hazy solution results. Regardless of the cloudiness, the enzyme is active and should be usable for metabolite hydrolysis.

Storage/Stability

The product, as supplied, should be stored at –20 °C.

A solution at ≥ 5 mg/mL in 75 mM phosphate buffer (pH 6.8) may be stored at –20 °C for up to 2 months with little or no loss of activity.

References

1. Kim, D.-H. *et al*., *Biol. Pharm. Bull*., **18(9)**, 1184-1188 (1995).
2. Jefferson, R.A. *et al*., *Proc. Natl. Acad. Sci. USA*, **83(22)**, 8447-8451 (1986).
3. Tephly, T.R. *et al*., *Adv. Pharmacol.*, **42**, 343-346 (1998).
4. Bergmeyer, H.-U. (ed.), *Methods of Enzymatic Analysis*, 2nd edition. Academic Press (New York, NY), pp. 460-461, 929-943 (1974).
5. Graef, V. *et al*., *Clin. Chem*., **23(3)**, 532-535 (1977).
6. Tsujikawa, K. *et al*., *Journal of Health Science*, **50(3)**, 286-289 (2004).
7. Meatherall, R., *J. Anal. Toxicol*., **18(7)**, 382-384 (1994).
8. Burnet, J.-B. *et al*., *Water Res*., **164**, 114869 (2019).
9. Satoh, H. *et al*., *Sci. Total Environ*., **715**, 136928 (2020).
10. Kim, J.H. *et al*., *J. Microbiol. Biotechnol*., **31(11)**, 1576-1582 (2021).
11. Medina-Pérez, G. *et al*., *Appl. Sci*., **11(16)**, 7705 (2021).
12. Yue, B. *et al*., *Front. Pharmacol*., **12**, 774560 (2021).

Notice

We provide information and advice to our customers on application technologies and regulatory matters to the best of our knowledge and ability, but without obligation or liability. Existing laws and regulations are to be observed in all cases by our customers. This also applies in respect to any rights of third parties. Our information and advice do not relieve our customers of their own responsibility for checking the suitability of our products for the envisaged purpose.

The information in this document is subject to change without notice and should not be construed as a commitment by the manufacturing or selling entity, or an affiliate. We assume no responsibility for any errors that may appear in this document.

Technical Assistance

Visit the tech service page at SigmaAldrich.com/techservice.

Standard Warranty

The applicable warranty for the products listed in this publication may be found at SigmaAldrich.com/terms.

Contact Information

For the location of the office nearest you, go to SigmaAldrich.com/offices.

The life science business of Merck operates as MilliporeSigma in the U.S. and Canada.

Merck and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates. All other trademarks are the property of their respective owners. Detailed information on trademarks is available via publicly accessible resources.

© 2022 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.

G7396dat Rev 04/22 AC,GCY,MAM