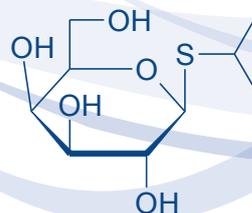


IPTG

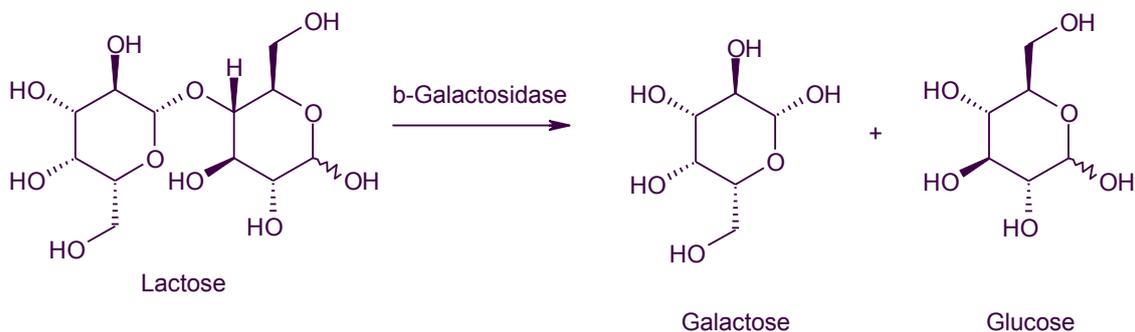
Product Code: **EI05931**
 CAS Number: **367-93-1**
 Chemical Formula: **C₉H₁₈O₅S**
 Molecular Weight: **238.3**



Synonyms: *Isopropyl β-D-thiogalactopyranoside*
IPTG non-animal origin

IPTG is the usual abbreviation for isopropyl β-D-thiogalactopyranoside. It is typically produced using D-galactose derived from milk lactose. Carbosynth also manufactures a grade that is of plant origin and therefore does not face the usual TSE/BSE issues. We call this grade “IPTG non-animal origin” and it is available as a free flowing crystalline powder in a range of packaging from 5 g to 20 kg drums.

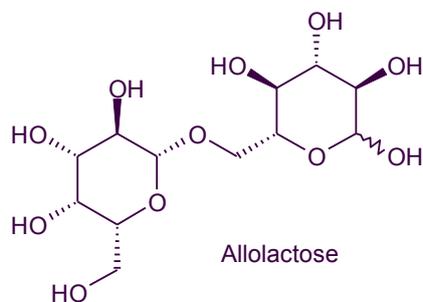
IPTG is used as a molecular mimic of allolactose, a lactose metabolite that promotes transcription of the lac operon¹. The lac operon is the regulated group of genes required for the transport and metabolism of lactose in *Escherichia coli*. It contains three adjacent genes, lacZ, lacY and lacA. lacZ encodes β-galactosidase, an hydrolase enzyme that cleaves the disaccharide lactose into glucose and galactose to provide energy, Scheme 1.



Scheme 1

The lac operon is regulated by several factors including the availability of glucose and lactose. Gene regulation of the lac operon was the first complex genetic regulatory mechanism to be elucidated and is one of the foremost examples of prokaryotic gene regulation.

Production of β-galactosidase when no lactose is available to the cell, or when glucose is available as an alternative energy source, would be unnecessary and inefficient. Therefore the lac operon is controlled to ensure that β-galactosidase and other enzymes necessary for lactose metabolism are only produced when appropriate. In the absence of lactose a regulatory protein, lac repressor, prevents production of β-galactosidase by binding tightly to a short DNA sequence in the lac operon called the lac operator. Binding of the lac repressor inhibits binding of the enzyme RNA polymerase to the DNA, thus preventing its transcription. When lactose is available to the cells, the metabolite allolactose (an isomeric form of lactose) binds to the lac repressor releasing the operator and allowing transcription of the lac genes by RNA polymerase and hence production of the encoded proteins.



Scheme 2.

E. coli is one of the most widely used hosts for recombinant protein expression. The gene *lac Z* can be replaced with another gene coding for a protein of interest and the *lac* operator activated by IPTG for protein production^{2, 3}.

IPTG is effective as an inducer of the *lac* operon by acting as a mimic of allolactose. It binds to the *lac* repressor freeing the *lac* operon for transcription. IPTG is advantageous for *in vivo* studies as, unlike allolactose, the sulfur linkage is non-hydrolyzable by *E. coli*, preventing degradation of the inducer and hence its concentration remains constant and the rate of expression of *lac p/o*-controlled genes is non-variable in the experiment. IPTG is typically an effective inducer in the concentration range 100 μ M to 2mM. For induction, a sterile 1 M solution of IPTG is typically added by 1:1000 dilution into a logarithmically growing bacterial culture although higher concentrations may be used.

X-Gal (5-Bromo-4-chloro-3-indolyl β -D-galactopyranoside; Carbosynth product code EB06680) is frequently used in molecular biology applications to indicate the activity of β -galactosidase. X-Gal is cleaved by the enzyme to yield galactose and 5-bromo-4-chloro-3-hydroxyindole, which then oxidises into an insoluble blue product. X-Gal and IPTG are used in combination on an agar medium on a culture plate, and bacterial colonies with a functional *lacZ* gene can be readily identified by their blue colour.⁴

References:

1. Hansen, L. H.; Knudsen, S.; Sørensen, S. J. *Curr. Microbiol.* **1998**, *36*, 341.
2. Goeddel, D. V.; Kleid, D. G.; Bolivar, F.; Heyneker, H. L.; Yansura, D. G.; Crea, R.; Hirose, T.; Kraszewski, A.; Itakura, K.; Riggs, A. D. *Proc. Natl. Acad. Sci. USA* **1979**, *76*, 106.
3. Itakura, K.; Hirose, T.; Crea, R.; Riggs, A. D.; Heyneker, H. L.; Bolivar, F.; Boyer, H. W. *Science* **1977**, *198*, 1056.
4. Sambrook, J.; Russell, D. W. *Molecular Cloning: A Laboratory Manual*, 3rd Edition, Cold Spring Harbor Laboratory Press, 2001.