

Product No. 07663

Zymolyase™-20T (from *Arthrobacter luteus*)

Source: *Arthrobacter luteus*

Description:

Zymolyase™-20T, produced by a submerged culture of *Arthrobacter luteus*¹, is a new enzyme preparation which lyses effectively cell walls of viable yeast cells^{2, 3}. This preparation is a lyophilized powder prepared by salting out from culture fluid with ammonium sulfate.

An essential enzyme responsible for lysis of viable yeast cells in this preparation is β -1,3-glucan laminaripentaohydrolase. It hydrolyzes linear glucose polymers with β -1,3-linkages and releases specifically laminaripentaose as the main and minimum product unit^{4,5,10,11}.

The extent of lysis of yeast cells by Zymolyase™-20T varies with yeast strain, growth stage of yeast, or cultural condition⁶⁻⁸.

Zymolyase™-20T shows 20,000 units/g of the lytic activity, defined after, toward brewer's yeast cells (*Saccharomyces cerevisiae*, resting stage) or toward yeast cells of *Saccharomyces cerevisiae* IFO 0565 cultured statically in malt extract medium (malt extract 2g, peptone 0.5g, water 100ml) at 20°C for 34hr.

Further purified preparation⁹ is also available as Zymolyase™-100T whose specific activity is 100,000units/g.

Further informations related to Zymolyase™ are obtained in the references cited below¹²⁻¹⁶.

Product information:

Activity	20,000 units/g	
Contaminants	β -1, 3-glucanase	1.5×10^6 units/g
	Protease	1.0×10^4 units/g
	Mannanase	1.0×10^6 units/g
	(See reference No.3 as to the definition of each enzyme units. Each activity varies more or less amount lots.)	
	Amylase, Xylanase, Phosphatase	Minute amounts
Essential Enzyme	β -1, 3-glucan laminaripentaohydrolase	
Appearance	Lyophilized powder	
Optimum pH and temperature	pH7.5, 35°C (for lysis of viable yeast cells)	
	pH6.5, 45°C (for hydrolysis of yeast glucan)	
Stable pH	5-10	
Heat stability	The lytic activity is lost on incubation at 60°C for 5 minutes.	
Specificity (Lytic spectrum) ⁵	<i>Ashbya</i> , <i>Candida</i> , <i>Debaryomyces</i> , <i>Eremothecium</i> , <i>Endomyces</i> , <i>Hansenula</i> , <i>Hanseniaspora</i> , <i>Kloeckera</i> , <i>Kluyveromyces</i> , <i>Lipomyces</i> , <i>Metschnikowia</i> , <i>Pichia</i> , <i>Pullularia</i> , <i>Torulopsis</i> , <i>Saccharomyces</i> , <i>Saccharomycopsis</i> , <i>Saccharomycodes</i> , <i>Schwanniomyces</i> , etc.	
Activator	SH compound such as cysteine, 2-mercaptoethanol or dithiothreitol	

Unit Definition:

One unit of lytic activity is defined as that amount which indicates 30% of decrease in absorbance at 800nm (A_{800}) of the reaction mixture under the following condition.

Assay for Enzyme Activity:

Method

[Reaction mixture]

Substrate and Buffer solution:	Brewer's yeast cell suspension (2mg dry weight/ml)	3 mL
	M/15 Phosphate buffer, pH7.5	5 mL
Enzyme solution:	0.05-0.1 mg/mL solution	1 mL
Distilled water		1 mL
Total volume		10 mL

[Procedure]

After incubation for 2 hours at 25°C with gentle shaking, A₈₀₀ of the mixture is determined. As a reference, 1 mL of distilled water is used instead of enzyme solution.

Calculation

Percentage decrease in A₈₀₀ = (A₈₀₀ of reference – A₈₀₀ of reaction mixture) × 100/ initial A₈₀₀ of reference

When 60% of A₈₀₀ decrease, equivalent to 2 units, is observed in the reaction system, the brewer's yeast cells are completely lysed, namely, 1 unit of Zymolyase™-20T lyses 3mg dry weight of brewer's yeast.

Precautions on use:

Use a sterilized filter except nitrocellulose when a sterilized enzyme solution is needed.

Storage:

Stable for at least 1 year at 2°C. About 70% of the lytic activity is lost when stored at 30°C for 3 months.

References:

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- 5) Kaneko, T., Kitamura, K. and Yamamoto, Y.: *Agric. Biol. Chem.*, **37**, 2295(1973)
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Note: For *in vitro* research use only, not for diagnostic or therapeutic use. This product is not a medical device.

Manufacturer : Mitsubishi Corporation Life Sciences Limited

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