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07663E_1109_0

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 condition[6-8].

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 preparation[9] is also available as ZYMOLYASE®-100T whose specific activity is 100,000units/g.

Further informations related to ZYMOLYASE® are obtained in the references sited below[12-16].

Specifications:

Activity	20,000units/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]	*Ashbya, Candida, Debaryomyces, Eremothecium, Endomyces, Hansenula, Hanseniaspora, Kloekera, Kluyveromyces, Lipomyces, Metschnikowia, Pichia, Pullularia, Torulopsis, Saccharomyces, Saccharomycopsis, Saccharomycodes, Schwanniomyces*, 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.

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Assay for Enzyme Activity:

Method

[Reaction mixture]

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

[Procedure]

After incubation for 2 hours at 25°C with gentle shaking, A_{800} of the mixture is determined. As a reference,1ml of distilled water is used instead of enzyme solution.

Calculation

Percentage decrease in A_{800} = (A_{800} of reference – A_{800} of reaction mixture) × 100/ initial A_{800} of reference

When 60% of A_{800} 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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Note: For *in vitro* research use only, not for diagnostic or therapeutic use. This product is not a medical device.

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