

#### Yeast Protein Kit™

Simple and convenient method for the rapid and thorough lysis of yeast cells.

#### **Highlights**

- · Convenient method to lyse yeast cell for protein and PCR analysis.
- · The kit can be used for any fungus species that are susceptible to yeast lytic enzyme digestion. It is optimized for S. cerevisiae and C. albicans.

Catalog Numbers: Y1002



Scan with your smart-phone camera to view the online protocol/video.





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## **Product Contents**

Yeast Protein Kit™	<b>Y1002</b> (200 Preps.)	Storage Temperature
Y-Lysis Buffer	6 ml	Room Temp.
Zymolyase <sup>TM</sup> , (Resuspend the lyophilized enzyme by adding 200 μl of <b>Storage Buffer</b> to make 5 units/μl)	1000 U	Shipped at room temperature. Store at -20°C after arrival.
Instruction Manual	1	-

### **Product Description**

The **Yeast Protein Kit™** is a simple and convenient method to lyse yeast cells for protein analysis, such as Western Blot Analysis, and PCR amplification of plasmid or genomic DNA. The kit essentially generates spheroplasts of yeast cells for subsequent analysis. It is optimized for *S. cerevisiae* and *C. albicans*. The kit can also be used for any fungus species that is susceptible to yeast lytic enzyme digestion.

#### **Protocol**

#### **Zymolyase Preparation**

<u>Before starting</u>: Add 200 µl of the supplied **Storage Buffer** to the lyophilized **Zymolyase™**. Mix to dissolve the enzyme completely, spin briefly in a micro-centrifuge. Store the reconstituted **Zymolyase™** at -20°C.

#### **Western Blot Analysis**

This method produces spheroplasts of yeast cells. Fresh and log-phase cells are more susceptible to **Zymolyase™**. Try to use fresh cells whenever possible.

1. Spin down 1 x 10<sup>5-6</sup> cells and remove the supernatant.

Use 200-500  $\mu$ l yeast liquid culture in most situation. Remove supernatant as much as possible.

2. Add 25 µl of **Y-Lysis Buffer** and 1 µl of **Zymolyase**™ to the sample.

For multiple sample analysis, add 40  $\mu$ l of **Zymolyase<sup>TM</sup>** to 1 ml of **Y-Lysis Buffer**. Use 25  $\mu$ l of this mixture for each sample.

- Incubate at 37°C for 30-60 minutes.
- 4a. Add 25 μl of 2X SDS-PAGE sample buffer to the digested yeast cell suspension and the sample can be used directly for SDS\_PAGE analysis. If the experiment requires remove of the Y-Lysis Buffer, use procedure in step 4b below.
- 4b. Centrifuge the cell at 500 g for 5 minutes and remove the supernatant. The pellet can be directly resuspended in SDS-PAGE sample buffer for gel analysis.

#### **PCR Analysis**

Two protocols are provided. In **Protocol I**, simply add 0.5 -1 µl of **Zymolyase™** directly to each PCR reaction (20-100 µl reaction) plus yeast cells. **Protocol II** makes yeast spheroplast first, and then the spheroplast is used as a template for PCR analysis. Both methods work.

#### **PCR Protocol I**

- 1. Remove a small amount of yeast cells, ~0.1-0.5 μl from a colony or pellet and add it directly to the PCR reaction.
- Add 0.5 µl of Zymolyase<sup>™</sup> to each 20-100 µl of PCR reaction. Proceed to PCR directly.

If the PCR is set-up at room temperature, proceed directly to PCR reaction. If the PCR is set-up on ice, incubate sample at 37°C for 5 minutes before starting the PCR reaction.

#### PCR Protocol II

This method is suitable for both liquid cultures and colonies.

1. Pellet approximately 1 x  $10^{6-7}$  yeast cells and remove the supernatant.

For colonies, pick a small amount of yeast cells,  $\sim$ 0.5-2  $\mu$ l volume, using a pipette tip and dispense into the lysis solution in Step 2 below.

 Add 20 µl of Y-Lysis Buffer and 1 µl of Zymolyase™ to the sample.

For multiple sample analysis, add 40 µl of **Zymolyase™** to 1 ml of **Y-Lysis Buffer**. Use 25 µl of this mixture for each sample.

Incubate at 37°C for 5-10 minutes.

The cell suspension is ready for PCR reactions. Use 2-4  $\mu$ l of this digested cell suspension for each PCR reaction. The remaining portion can be frozen for future use.

# **Ordering Information**

Product Description	Catalog No.	Size
Yeast Protein Kit™	Y1002	200 Preps.

Individual Kit Components	Catalog No.	Amount
Y-Lysis Buffer	Y1002-1-6	6 ml
<b>Zymolyase</b> ™ (with Zymolyase Storage Buffer)	E1004	1000 Units (lyophilized), Storage Buffer 500 μl



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Integrity of kit components is guaranteed for up to one year from date of purchase. Reagents are routinely tested on a lot-to-lot basis to ensure they provide the highest performance and reliability.

This product is for research use only and should only be used by trained professionals. It is not for use in diagnostic procedures. Some reagents included with this kit are irritants. Wear protective gloves and eye protection. Follow the safety guidelines and rules enacted by your research institution or facility.

<sup>&</sup>lt;sup>™</sup> Trademarks of Zymo Research Corporation Zymolyase<sup>™</sup> is a trademark of the Kirin Brewery Co., Ltd



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