

Yeast DNA Preparation - Solution Kit

Cat#: orb533443 (Datasheet)

Yeast DNA Preparation - Solution Kit

Solution based genomic DNA purification from yeast

For *in vitro* use only!

Shipping: shipped at ambient temperature

Storage Conditions: store at ambient temperature

Shelf Life: 12 months

Description:

Yeast DNA Preparation Kit is designed for convenient and fast isolation of genomic DNA from yeast cells. The solution based system minimizes DNA fragmentation that may be problematic in other spin-column/filtration based method. Because phenol or chloroform is not used it is safe and does not produce any harmful waste. Solution based genomic DNA purification kits guarantee minimal DNA fragmentation and yield DNA sized up to 150 kb.

Expected yield:

Yields of genomic DNA will vary from sample to sample depending on the amount, quality and type of material processed. An amount of approx. 10 µg purified DNA per preparation can be expected.

Content:

Cell Resuspension Solution

Lyticase (before use, solve in Lyticase Suspension Solution to obtain a final concentration of 2.5 units/µl) - store at -20 °C

Lyticase Suspension Solution

Cell Lysis Solution

Protein Precipitation Solution

Washing Buffer (before use, add 96-99 % Ethanol as indicated on the bottle)

DNA Hydration Solution RNase A (before use, solve in double distilled water to obtain a final concentration of 4 mg/ml) - store at -20 °C

To be provided by you:

Isopropanol (2-propanol) >99 %

96-99 % Ethanol

Microtubes 1.5 ml

Preparation procedure:

Before start, provide >99 % Isopropanol (2-propanol) (not included in the kit).

For S pack (100 preps): Add 120 µl Lyticase Suspension Solution to the Lyticase tube, 200 µl dd-water to the RNase A tube and 48 ml 96-99 % Ethanol (not included in the kit) to the Washing Buffer bottle.

For L pack (500 preps): Add 120 µl Lyticase Suspension Solution to each Lyticase tube, 200 µl dd-water to each RNase A tube and 120 ml 96-99 % Ethanol (not included in the kit) to each Washing Buffer bottle.

Buffer	100 preps	500 preps
Cell Resuspension Solution	32 ml	160 ml
Lyticase (2.5 units/µl)	300 units	5x 300 units
Lyticase Suspension Solution	130 µl	650 µl
Cell Lysis Solution	32 ml	160 ml
Protein Precipitation Solution	11 ml	55 ml
Washing Buffer	add 48 ml Ethanol (final volume 60 ml)	add 120 ml Ethanol to each bottle (final volume 150 ml each)
DNA Hydration Solution	11 ml	55 ml
RNase A (4 mg/ml)	0.8 mg	5x 0.8 mg

1 Cell Lysis:

- Transfer 1 ml of cultured cells into a 1.5 ml microtube
- Harvest the cells by centrifuging at 15,000 g for 1 min and discard the supernatant

- Resuspend the cell pellet in 300 µl of Cell Resuspension Solution
- Add 1 µl of Lyticase Solution and mix by inverting approx. 25 times
- Place the tube at 37 °C for 30-60 min
- Centrifuge at 15,000 g for 1 min and discard the supernatant
- Resuspend the pellet in 300 µl of Cell Lysis Solution

2 Protein Precipitation:

- Add 100 µl of Protein Precipitation Solution and vortex vigorously for 20 sec
- Centrifuge at 15,000 g for 5 min

3 DNA Precipitation:

- Pour the supernatant to a clean 1.5 ml microtube containing 300 µl Isopropanol >99 %
- Mix the sample by inverting gently 50 times
- Centrifuge at 15,000 g for 1 min (DNA should be visible as a small white pellet)
- Discard the supernatant and drain tube briefly on clean absorbent paper. Add 500 µl Washing Buffer and invert the tube several times to wash the DNA pellet
- Centrifuge at 15,000 g for 1 min. Discard the ethanol carefully.
- Air dry at room temperature for 10-15 min

4 DNA Hydration:

- Add 50-100 µl of DNA Hydration Solution to the dried DNA pellet
- Add 1.5 µl of RNase A Solution and incubate at 37 °C for 30 min
- Hydrate the DNA by incubating for 60 min at 65 °C
- Store the DNA at 4 °C. For long time storage, place sample at -20 °C or -80 °C