

Animal and Fungi DNA Preparation Kit

DNA purification from animal tissue and fungi

Solution based genomic DNA Preparation Kit

| | Cat.-No. | Amount |
|--|----------|------------------|
| | PP-208XS | 20 preparations |
| | PP-208S | 100 preparations |
| | PP-208L | 500 preparations |

For *in vitro* use only.

Quality guaranteed for 12 months.

Store at room temperature.

For long term storage place *Proteinase K* and *RNase A lyophilisates* at -20°C.

Proteinase K and *RNase A Solutions* should be stored at -20°C.

Kit contents

Cell Lysis Solution

Cell Resuspension Solution

Proteinase K (before use, solve in double distilled water to obtain a final concentration of 20 mg/ml)

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)

To be provided by you

Isopropanol (2-propanol) >99%

96-99% Ethanol

Microtubes 1.5 ml

Description

Animal and Fungi DNA Preparation Kit is designed for convenient and fast isolation of genomic DNA from animal tissue and fungi. The solution based system minimizes DNA fragmentation that may be problematic in spin-column / filtration based methods. 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.

| Buffer | PP-208XS | PP-208S | PP-208L |
|--------------------------------|---|---|---|
| | 20 preps | 100 preps | 500 preps |
| Cell Lysis Solution | 6.4 ml | 32 ml | 160 ml |
| Cell Resuspension Solution | 6.4 ml | 32 ml | 160 ml |
| <i>Proteinase K</i> | 0.8 mg (before use, solve in 40 µl dd-water to obtain a final concentration of 20 mg/ml) | 4 mg (before use, solve in 200µl dd-water to obtain a final concentration of 20 mg/ml) | 5 x 4 mg (before use, solve each vial in 200 µl dd-water to obtain a final concentration of 20 mg/ml) |
| Protein Precipitation Solution | 2.2 ml | 11 ml | 55 ml |
| Washing Buffer | add 9.6 ml Ethanol (final volume 12 ml) | add 48 ml Ethanol (final volume 60 ml) | add 120 ml Ethanol to each bottle (final volume 150 ml each) |
| DNA Hydration Solution | 2.2 ml | 11 ml | 55 ml |
| <i>RNase A</i> | 0.16 mg (before use, solve in 40 µl dd-water to obtain a final concentration of 4 mg/ml) | 0.8 mg (before use, solve in 200 µl dd-water to obtain a final concentration of 4 mg/ml) | 5 x 0.8 mg (before use, solve each vial in 200 µl dd-water to obtain a final concentration of 4 mg/ml) |

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Preparation procedure

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

For XS pack (20 preps): Add 40 µl dd-water to the *Proteinase K* tube, 40 µl dd-water to the *RNase A* tube and 9.6 ml 96-99% Ethanol (not included in the kit) to the *Washing Buffer* bottle.

For S pack (100 preps): Add 200 µl dd-water to the *Proteinase K* 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 200 µl dd-water to each *Proteinase K* 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.

Proteinase K and *RNase A* Solutions should be stored at -20°C.

1a Cell Lysis for Animal Tissue

- Transfer 5-10 mg of fresh or frozen tissue to a 1.5 ml microtube.
- Add 300 µl *Cell Lysis Solution* to the tissue.
- Add 1.5 µl *Proteinase K Solution* to the lysate and mix by inverting several times.
- Incubate at 55°C overnight or until tissue has dissolved.

1b Cell Lysis for Fungi

- Transfer 1 ml of the cultured cells to a 1.5 ml microtube.
- Harvest the cells by centrifuging at 15,000 g for 1 min and discard supernatant.
- Resuspend the cell pellet in 300 µl *Cell Resuspension Solution*.
- Add 1.5 µl *Proteinase K Solution* and mix by inverting several times.
- Incubate at 55°C for 60 min.
- Centrifuging at 15,000 g for 1 min and discard supernatant.
- Resuspend the pellet in 300 µl *Cell Lysis Solution*.

2. Protein Precipitation

- Add 100 µl of *Protein Precipitation Solution* to the cell lysate.

- Mix the solution well by vortexing for 20 sec.
- Centrifuge at 15,000 g for 3 min. (The precipitated protein will be a tight pellet. If the pellet is not tight, repeat mixing, incubate on ice for 10 minutes, and then centrifuge again.)

3. DNA Precipitation

- Transfer the supernatant to a clean 1.5 ml microtube containing 300 µl of *Isopropanol* >99%.
- Mix the sample by inverting gently 50 times.
- Centrifuge at 15,000 g for 1 min. The DNA will be visible as a pellet that ranges in color from off-white to light green.
- Discard the supernatant and drain tube briefly on clean absorbent paper.
- Add 500 µl *Washing Buffer* and invert 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* to the cell lysate.
- Mix the sample by inverting the tube and incubate at 37°C for 30-60 min.
- Hydrate the DNA by incubating sample at 65°C for 60 min.
- Store DNA at 4°C. For long time storage, place sample at -20°C or -80°C.