A&A BIOTECHNOLOGY
innovating life science

# Genomic Mini

Versatile kit for genomic DNA purification from various sources.

**version 0517**

## 50 isolations, 250 isolations

Cat. # 116-50, 116-250

Kit for DNA isolations from z tissues, bacteria, cell culture.
The binding capacity of the DNA purification column is 20 µg of DNA.

For R&D use only

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## Kit Contents

| Component | 50 isolations | 250 isolations | Store at |
|---|---|---|---|
| Minicolumns | 50 pcs | 250 pcs | Room Temp. |
| 2 ml tubes | 50 pcs | 250 pcs | Room Temp. |
| A1 wash solution | 50 ml | 250 ml | Room Temp. |
| LT lysis solution<br>**Note:** If LT solution contains the precipitate, heat it to 40 °C until dissolution of complete precipitate. | 15 ml | 60 ml | Room Temp. |
| Tris buffer (10 mM, pH 8,5) | 25 ml | 125 ml | Room Temp. |
| Proteinase K | 1.1 ml | 5 x 1.1 ml | +4 to +8 °C |

## Equipment and materials necessary for the DNA isolation that are not included in kit

1. Material for DNA isolation
2. Enzymes (option – depending on type of biological material):
   Lysozyme – 10 mg/ml, 400 U/µl (cat. # 1005-10, 1005-50)
   Lysostaphin – 0.4 U/µl (cat. # 1007-400, 1007-2000)
   Mutanolysin – 10 U/µl (cat. # 1017-5, 1017-10, 1017-50)
3. RNAse – 10 mg/ml (cat. # 1006-10, 1006-50) (optional)
4. Sterile water (nuclease free, DEPC treated) (cat. # 003-075, 003-25) (optional)
5. DTT (dithiothreitol) (cat. # 2010-5, 2010-25, 2010-10P) (optional)
6. 1.5 ml Eppendorf tubes
7. Incubator or heatblock set to 37 °C, 50 °C, 70 °C
8. Vortex
9. Microentrifuge

**NOTE:**

Before you start working, we recommend cleaning the work surface using LabZAP™ product (cat. # 040-500).

A&A Biotechnology provides one year guarantee on this kit

The company does not guarantee correct performance of this kit in the event of:

- not adhering to the supplied protocol
- not recommended use of equipment and materials
- the use of other reagents than recommended or which are not a component of the kit
- the use of expired or improperly stored reagents and minicolumns

# Material preparation

## Bacteria

1. Transfer 100 µl of bacterial culture to a 1.5 ml tube (not included).
   **NOTE:** For bacterial culture 200 µl–1 ml volume: centrifuge the sample, discard supernatant, suspend the bacterial pellet in 100 µl of Tris buffer.
2. For Gram(+) bacteria, we recommend using the following enzymes:

   for *S.aureus* we recommend using lysostaphin (0.4 U/µl)
   (not included, cat # 1007–400, 1007–2000);
   add 10 µl of lysostaphin and incubate for 10 min at 37 °C.

   for *Streptococcus, Lactobacillus, Lactococcus, Listeria* we recommend using mutanolysin (10 U /µl) (not included, cat. # 1017–5, 1017–10, 1017–50) or mutanolysin with lysozyme (not included, cat # 1005–10, 1005–50);
   add 5 µl of mutanolysin or 5 µl of mutanolysin and 10 µl of lysozyme.
   Mix and incubate for 20 min at 50 °C.

3. Add 200 µl of LT lysis solution and 20 µl of Proteinase K solution.
4. Mix the whole sample by inverting the tube and incubate for 20 min at 37 °C.
   **RNA digestion (optional):** Add 5 µl of RNAse (10 mg/ml solution)
   (not included, cat. # 1006–10, 1006–50). Mix sample and incubate for 5 min at room temp.
5. Follow point 1. of the protocol

## Semen

1. Transfer 100 µl of semen to a 1.5 ml tube (not included).
2. Add 10 µl of 1M DTT (not included, cat # 2010–5, 2010–25, 2010–10P.
3. Add 200 µl of LT lysis solution and 20 µl of Proteinase K solution.
4. Mix the sample by inverting the tube and incubate fo 20 min at 37 °C.
   **RNA digestion (optional):** Add 5 µl of RNAse (10 mg/ml solution)
   (not included, cat. # 1006–10, 1006–50). Mix sample and incubate for 5 min at room temp.
5. Follow point 1. of the protocol

## Cell culture

1. Transfer $1x10^6$ of cell culture to a 1.5 ml tube (not included). Centrifuge and discard the supernatant.
2. Suspend the pellet in 100 µl of Tris buffer.
3. Add 200 µl of LT lysis solution and 20 µl of Proteinase K solution.
4. Mix the sample by inverting the tube and incubate for 20 min at 37 °C.

   **RNA digestion (optional):** Add 5 µl of RNAse (10 mg/ml solution) (not included, cat. # 1006-10, 1006-50). Mix sample and incubate for 5 min at room temp.
5. Follow point 1. of the protocol

## Fresh tissues

1. Up to 10-15 mg of fragmented tissues transfer to a 1.5 ml tube (not included).
2. Add 100 µl of Tris buffer, 50 µl of LT lysis solution and 20 µl of Proteinase K solution.
3. Mix the sample by vortexing. Incubate at 50 °C up to completly lyse of tissue. Mix the sample from time to time by vortexing.
4. Mix the sample by vigorous vortexing for 20 s.

   **RNA digestion (optional):** Add 5 µl of RNAse (10 mg/ml solution) (not included, cat. # 1006-10, 1006-50). Mix sample and incubate for 5 min at room temp.
5. Add 150 µl of LT lysis solution and mix the sample.
6. Follow point 1. of the protocol

## Embedded tissues

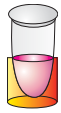
1. Transfer the tissue to a 1.5 ml tube (not included).
2. Add Tris buffer, centrifuge and discard supernatant. Repeat several times to remove the fixing liquid.
3. Remove the paraffin from FFPE samples - rinse in xylen (not included), next rinse in ethanol (not included).
4. Follow the fresh tissues of material preparation.

For embedded tissues we recommend Xpure™ FFPE micro (cat. # 091-50) -
Kit for genomic DNA purification from tissues preserved in paraffin.
Fast deparaffinization without xylene and hexane.

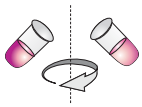
## Isolation protocol

1. Set the thermoblock temperature to 70 °C and place in it the tubes with Tris elution buffer (it will be used in point 8. of protocol).


2. Incubate the samples for 5 min at 70 °C.




3. Mix by vortexing for 20 s and centrifuge for 1 min at 10 000–15 000 RPM.

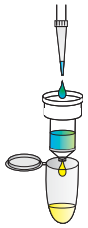


4. Apply the supernatants onto the minicolumns.

   Centrifuge for 1 min at 10 000–15 000 RPM.

5. Add 500 µl of A1 wash solution.

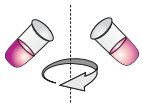
   Centrifuge for 1 min at 10 000–15 000 RPM.



6. Transfer the minicolumns to the new 2 ml tubes (included).

   Add 400 µl of A1 wash solution.

7. Centrifuge for 1 min at 10 000–15 000 RPM.



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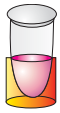
8. Transfer the minicolumns to the new 1.5 ml tubes (not included).

Add to the minicolumns 200 µl of Tris buffer (included) or sterile water (not included) heated to 70 °C.

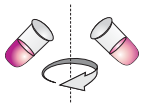
NOTE: If it is expected the low isolation yield, the elution volume (Tris buffer or water) can be reduced to 100 µl increasing the final DNA concentration.

9. Incubate the samples for 2 min at room temp.



room temp.

10. Centrifuge for 1 min at 10 000–15 000 RPM.



11. Remove the minicolumns.
Store the purified DNA at +4 °C or –20 °C.

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## Related products

| Product | Quantity | Cat. # |
|---|---|---|
| Xpure Genomic Mini | 50 isolations | 096-50 |
| Xpure Cell&Tissue | 50 isolations | 090-50 |
| Genomic Midi AX | 20 isolations | 895-20 |
| Genomic Midi AX Direct | 20 isolations | 895-20D |
| Genomic Maxi AX | 10 isolations | 995-10 |
| Genomic Maxi AX Direct | 10 isolations | 995-10D |
| Plasmid Mini | 50 isolations | 020-50 |
| | 250 isolations | 020-250 |
| Gel-Out | 50 isolations | 023-50 |
| | 250 isolations | 023-250 |
| Clean-Up | 50 isolations | 021-50 |
| | 250 isolations | 021-250 |
| ExTerminator | 50 isolations | 444-50 |
| | 250 isolations | 444-250 |
| Swab | 25 isolations | 025-25 |
| | 100 isolations | 025-100 |
| Genomic Mini AX Swab&Semen Spin | 100 isolations | 025-100S |

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# Safety information

**DANGER**

**Proteinase K**

H315 Causes skin irritation.
H319 Causes serious eye irritation.
H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.
H335 May cause respiratory irritation.
P261 Avoid breathing dust.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P342+P311 If experiencing respiratory symptoms call a Poison Center or doctor/physician.

**WARNING**

**LT lysis solution**

H302 Harmful if swallowed.
H315 Causes skin irritation.
H319 Causes serious eye irritation.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

**DANGER**

**A1 wash solution**

H225 Highly flammable liquid and vapour.
H319 Causes serious eye irritation.
H336 May cause drowsiness or dizziness.
P210 Keep away from heat, sparks, open flames, hot surfaces. No smoking.
P261 Avoid breathing vapours.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.