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MicroPrep DNA Purification Kit

For quick purification of total DNA in small elution volume.

DNA from enzymatic reactions, from agarose gels or plasmid DNA

Cat#: S5303

Version: 211018

GENAXXON bioscience

DNA Clean-up Protocol

1. **Add 2-10 volumes** of **CB Buffer** to 1 volume **of DNA sample** and vortex for 3 sec..

Application	Buffer to Sample ratio	Example
DNA fragments 100bp – 10kbp	5 : 1	500µL : 100µL
DNA fragments: <100bp	10 : 1	500µL : 50µL
Plasmid, genomic DNA	2 : 1	200µL : 100µL

NOTE: The solution should be yellow. If it turns pink after mixing, add 10µL of 3M sodium acetate, pH5.2, and mix thoroughly.
NOTE: For sample preparation method, see instructions given on page 3 (recommendations and important notes and page 4 PCR DNA Purification).

2. Centrifuge the tube briefly in order to recover any remaining liquid from the lid and transfer the whole volume of the mixture into a DNA purification column placed in a Collection Tube.
Centrifuge for 60 sec. at 11k-15k x g.
NOTE: If the volume of the mixture exceeds 800µL in total, discard the flow-through after centrifugation, then re-use the collection tube and transfer the remaining DNA sample mixture into the same MicroPrep purification column.
3. Transfer the DNA purification microPrep column to a new collection tube.
4. To wash, add **500µL CW Buffer** (add ethanol to **CW Buffer** before use) to the column and centrifuge for 30 sec. at 11k-15k x g.
5. Discard flow-through and re-use the Collection tube.
6. Add **500µL CW Buffer** and centrifuge for 30 sec. at 11k-15k x g.
7. Discard the flow-through and re-use Collection Tube.
8. Centrifuge for 60-120 sec. at 15k-21k x g.
NOTE: The wash buffer contains alcohol, which may interfere with some enzymatic reactions and also decrease elution efficiency. It is therefore vital to remove the alcohol completely from the column before elution.
9. Discard the collection tube with the flow-through and carefully transfer the microPrep purification column to a new sterile 1.5mL reaction tube.
10. Add **≥5µL Elution Buffer** directly onto the purification column membrane.
NOTE: It is essential to apply the elution buffer precisely onto the centre of the membrane.
NOTE: Other buffer volumes may be used. For instructions, see section: Recommendations and important notes on page 3.
11. Incubate the column at room temperature **for 120 sec**.
12. **Centrifuge at 11k-15k x g for 60 sec.**
13. Remove the MicroPrep purification column. The isolated DNA is ready for use in downstream applications or for short-term storage at +2°C to +8°C or for long-term storage at -20°C.

Gel extraction DNA isolation Protocol

1. Excise gel slice containing the DNA fragment and place it in a 1.5-2mL reaction tube
NOTE: The gel slice mass should not exceed 150mg. For instructions see page 4.
2. Add **500µL GB Buffer** and mix well by inverting the tube for several times.
3. Incubate the mixture at 50°C for 5-10 minutes or until the agarose has completely dissolved. During the incubation, mix the sample by inverting the tube several times.
NOTE: Ensure that the agarose is completely dissolved before moving on to the next step.
NOTE: The solution should be yellow. If it turns pink after mixing, add 10µL of 3M sodium acetate, pH5.2, and mix thoroughly.
4. Centrifuge the tube briefly to recover any remaining liquid from the lid and transfer **800µL of the mixture** into a DNA purification microPrep column placed in a Collection Tube.
Centrifuge for 1 min. at 11k-15k x g.
NOTE: If the volume of the mixture exceeds 800µL in total, discard the flow-through after centrifugation, then re-use the collection tube and transfer the remaining DNA sample mixture into the same MicroPrep purification column.
5. Transfer the DNA purification microPrep purification column to a new collection tube.
6. Add **500µL CW Buffer** (add ethanol to **CW Buffer** before use) to the column and centrifuge for 30 sec. at 11k-15k x g.
7. Discard flow-through and re-use the Collection tube.
8. Add **500µL CW Buffer** and centrifuge for 30 sec. at 11k-15k x g.
9. Discard the flow-through and re-use Collection Tube.
10. Centrifuge for 1-2 minutes at 15k-21k x g.
NOTE: The wash buffer contains alcohol, which may interfere with some enzymatic reactions and also decrease elution efficiency. It is therefore vital to remove the alcohol completely from the column before elution.
11. Discard the collection tube with the flow-through and carefully transfer the microPrep purification column to a new sterile 1.5mL reaction tube.
12. Add **≥5µL Elution Buffer** directly onto the purification column membrane.
NOTE: It is essential to apply the elution buffer precisely onto the centre of the membrane.
NOTE: Other buffer volumes may be used. For instructions, see section: Recommendations and important notes on page 3.
13. Incubate the column at room temperature **for 120 seconds (2 minutes)**.
14. **Centrifuge at 11k-15k x g for 60 seconds**
15. Remove the microPrep purification column. The isolated DNA is ready for use in downstream applications or for short-term storage at +2°C to +8°C or for long-term storage at -20°C.