



A Geno Technology, Inc. (USA) brand name

OmniPrep[™] Soil DNA

Isolate PCR Ready DNA from A Wide Variety of Environmental Samples

(Cat. # 786-469)



INTRODUCTION	3
ITEMS SUPPLIED	
STORAGE CONDITIONS	
IMPORTANT INFORMATION	
ADDITIONAL ITEMS REQUIRED	
PREPARATION BEFORE USE	
PROTOCOL	. 4
RELATED PRODUCTS	. 5

INTRODUCTION

The OmniPrep[™] Soil DNA kit provides all the reagents necessary to isolate PCR ready DNA for a large variety of environmental samples. The kit is primarily designed for use with environmental samples containing a high humic acid content, including difficult soil samples such as compost, manure and sediment. A major issue with high humic acid samples is the humic acids, and metals and polysaccharides, inhibit subsequent PCR. OmniPrep[™] Soil DNA SoilOUT[™] columns remove this interfering agents allowing for successful PCR.

ITEMS SUPPLIED (Cat. # 786-469)

Part. #	Description	Size
068G-C	Genomic Lysis Buffer	30ml
073L	LongLife Proteinase K (5mg/ml)	0.5ml
189D-B	DNA Stripping Solution	4 x 0.5ml
344P-B	Precipitation Solution	3 x 2ml
257S-A	SoilOUT [™] Columns	50
036T-A	TE Buffer	60ml

STORAGE CONDITIONS

The kit is shipped at ambient temperature. Upon arrival, store the kit components as recommended on the reagent label. This product is stable for 1 year at 4°C.

IMPORTANT INFORMATION

- Sample Source: Due to the wide variety of environmental samples compatible with this kit and the large variety of organic contaminants in these different samples a degree of optimization will be required. This will include the amount of sample used and volume of extract loaded on the SoilOUT[™] columns.
- Sample Homogenization: This kit uses a strong lysis buffer to release the DNA, however improved DNA yields may be achieved with bead mill homogenization, ultra-sonication or grinding the samples under liquid nitrogen.

ADDITIONAL ITEMS REQUIRED

- Isopropanol
- 70% ethanol
- 2ml collection tubes/ centrifuge tubes

PREPARATION BEFORE USE

Preheat water bath to 55-60°C

PROTOCOL

- Transfer 100mg soli sample to a 2ml centrifuge tube and add 400µl Genomic Lysis
 Buffer and 4µl LongLife[™] Proteinase K. Vigorously vortex to mix.
 NOTE: LongLife[™] Proteinase K is an enzyme suspension. Vortex vigorously before adding to the sample.
- 2. Incubate the sample at 55°C for 30-60 minutes with end-over-end mixing.
 NOTE: The OmniPrep[™] Soil DNA kit uses a strong lysis buffer to release the DNA, however improved DNA yields may be achieved with bead mill homogenization, ultra-sonication or grinding the samples under liquid nitrogen. We also recommend our EZ-Grind[™] product (Cat. # 786-139) for grinding the samples for improved DNA release.
- 3. Add 40µl DNA Stripping Solution and invert the tube several times to mix. Incubate the samples at 55°C for 10 minutes.
- 4. Centrifuge at 14,000xg for 5 minutes to pellet the solid debris. Transfer 250 μ l of the supernatant to a clean tube.
- Add 100μl Precipitation Solution and mix by inverting the tube several times. A
 white precipitate should be produced, if not add a further 50μl Precipitation
 Solution.

NOTE: In some cases the precipitate will be hard to see.

- 6. Centrifuge the sample at 14,000g for 10 minutes.
- 7. In the meantime, snap off the tab on the SoilOUT[™] column and place into a 2ml collection tube and centrifuge at 1,000xg for 2 minutes to remove the storage buffer. Add 0.5ml TE Buffer to the column and repeat the centrifugation. Discard the flow-through. Repeat the TE Buffer wash once more.
- 8. Place the column in a clean collection tube. Transfer 100μl supernatant from Step 6 to the SoilOUT[™] column and centrifuge at 1,000xg for 2 minutes. The flow-through contains the genomic DNA.
 - **NOTE:** For samples high in humic acid and other contaminants the volume added to the SoilOUT $^{\text{Tot}}$ column will need to be optimized. If a brown color passes through the column or inhibition of PCR occurs, load less onto the SoilOUT $^{\text{Tot}}$ column or use multiple columns.
- 9. Precipitate the DNA by adding 0.8 volumes isopropanol to the flow-through. Slowly invert the tube 10 times to precipitate the DNA.
- 10. Centrifuge at 14,000xg for 5 minutes to pellet the DNA. Remove and discard the supernatant.
- 11. Add 700µl 70% ethanol to the tube with the pellet and invert several times to wash excess salt from the pellet. Centrifuge at 14,000xg for 5 minutes to pellet the DNA NOTE: In some cases the pellet may be hard to see and be loosely attached to the tube. Take care when washing and removing supernatants.
- 12. Carefully decant or pipette off the 70% ethanol wash, invert the tube on a clean absorbent surface for several minutes to allow excess ethanol to drain off.

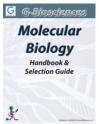
13. Add 50-100µl TE Buffer to the pellet and incubate at 55-60°C for at least 15 minutes to rehydrate the DNA.

NOTE: If required, RNase can be added at this stage to remove RNA.

14. The DNA is now ready for PCR amplification.

RELATED PRODUCTS

Download our Molecular Biology Handbook.



http://info.gbiosciences.com/complete-molecular-biology-handbook

For other related products, visit our website at <u>www.GBiosciences.com</u> or contact us.

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