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🌐 **QIAGEN DNeasy PowerSoil Kit**

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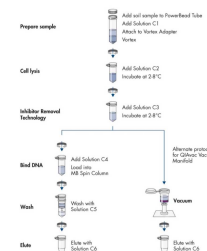
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Protocol status: Working

Protocol successful in several fish sedDNA studies

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Keywords: QIAGEN, DNeasy, PowerSoil, sedDNA, Sedimentary DNA, Fish, microbial genomic dna from all soil type, successful pcr amplification of organism, genomic dna from environmental sample, microbial genomic dna, isolated dna, isolating genomic dna, target fish species seddna, use with environmental sample, bacillus subtili, pcr amplification, successful pcr amplification, bacillus anthraci, including bacteria, pcr analysis, bacteria, containing high humic acid content, genomic dna, environmental sample, fungi, dneasy powersoil kit, dna, high humic acid content, including difficult soil type, organism, streptomyce, genomic, soil, algae, pcr, variety of organism, actinomycete, qiagen, difficult soil types such as compost, soil type, common soil

Abstract

For the isolation of microbial genomic DNA from all soil types

The DNeasy PowerSoil Kit comprises a novel and proprietary method for isolating genomic DNA from environmental samples using patented Inhibitor Removal Technology® (IRT). This kit is intended for use with environmental samples containing high humic acid content, including difficult soil types such as compost, sediment and manure. Other more common soil types have also been used successfully with this kit. The isolated DNA has a high level of purity, which allows for more successful PCR amplification of organisms from the sample. PCR analysis has been performed to detect a variety of organisms including bacteria (e.g., *Bacillus subtilis*, *Bacillus anthracis*), fungi (e.g., yeasts, molds), algae and actinomycetes (e.g., *Streptomyces*).

Protocol used successfully to detect target fish species sedDNA by Olajos et al., 2018; Nelson-Chorney et al., 2019; Thomson-Laing et al., 2020; Shiragaki et al., 2021; and Naro-Maciel et al., 2022

Materials

DNeasy PowerSoil Kit (50 preparations) already includes:

MB Spin Columns * 50

PowerBead Tubes * 50

Solution C1  6.6 mL

Solution C2  15 mL

Solution C3  15 mL

Solution C4  72 mL

Solution C5  30 mL

Solution C6  9 mL

Collection Tubes (2ml) 4 * 50

Quick Start Protocol

Additional equipments and reagents to be supplied by user:

Microcentrifuge ( 10.000 x g)

Pipettors (50 µl–500 µl)

Vortex-Genie 2 Vortex

Vortex Adapter for 24 (1.5–2.0 ml) tubes (cat. no. 13000-V1-24)

100% ethanol (for the QIAvac 24 Plus Manifold protocol only)

QIAvac 24 Plus Manifold

Safety warnings

-  Solution C5 contains ethanol. It is flammable.
Do not use bleach to clean the inside of the QIAvac® 24 Plus manifold.

Before start

Perform all centrifugation steps at room temperature (15–25°C).

If Solution C1 has precipitated, heat at 60°C until precipitate dissolves.



Sample preparation & cell lysis

1 **ADD**  0.25 g of soil sample to the PowerBead Tube

VORTEX gently to mix

2 **ADD**  60 μL of Solution C1

INVERT several times or vortex briefly

3 **SECURE** PowerBead Tubes horizontally using a Vortex Adapter for 24 (1.5–2.0 ml) tubes

10m

VORTEX at maximum speed for  00:10:00

4 **CENTRIFUGE** tubes at  10.000 x g for  00:00:30

30s

TRANSFER supernatant to a clean 2 mL Collection Tube

Inhibitor removal

25m 40s

5 **ADD**  250 μL of Solution C2

5m 5s

VORTEX for  00:00:05

INCUBATE at  2 °C to  8 °C for  00:05:00

6 **CENTRIFUGE** tubes at  10000 x g for  00:01:00

1m

AVOIDING the pellet, transfer up to  600 μL of supernatant to a clean 2 mL Collection Tube

7 **ADD**  200 μL of Solution C3

5m

VORTEX briefly to mix

INCUBATE at  2 °C to  8 °C for  00:05:00

8 **CENTRIFUGE** tubes at  10000 x g for  00:01:00

1m



AVOIDING the pellet, transfer up to  750 µL of supernatant to a clean 2 mL Collection Tube

Bind DNA

3m 5s

9 **SHAKE** to mix Solution C4

5s

ADD  1200 µL of Solution C4 to the supernatant

VORTEX for  00:00:05

10 **LOAD**  675 µL onto an MB Spin Column

1m

CENTRIFUGE at  10.000 x g for  00:01:00

DISCARD liquid flow-through

11 **REPEAT** step 10 twice, until all of the sample has been processed

Wash spin column

3m 5s

12 **ADD**  500 µL of Solution C5

30s

CENTRIFUGE at  10.000 x g for  00:00:30

DISCARD liquid flow-through

13 **CENTRIFUGE** again at  10.000 x g  00:01:00

1m

CAREFULLY place the MB Spin Column into a clean 2 mL Collection Tube. Avoid splashing any residual Solution C5 onto the column

Elute the DNA

3m 5s

14 **ADD**  100 µL of Solution C6 to the center of the white filter membrane

30s

CENTRIFUGE at  10000 x g for  00:00:30



DISCARD the MB Spin Column

DNA is now ready for downstream applications

Protocol references

<https://www.qiagen.com/us/resources/resourcedetail?id=5a0517a7-711d-4085-8a28-2bb25fab828a&lang=en>

Successful Publications:

Olajos, F., Bokma, F., Bartels, P., Myrstener, E., Rydberg, J., Öhlund, G., Bindler, R., Wang, X. R., Zale, R., & Englund, G. (2018). Estimating species colonization dates using DNA in lake sediment. *Methods in Ecology and Evolution*, 9(3), 535-543. <https://doi.org/10.1111/2041-210X.12890>

Nelson-Chorney, H. T., Davis, C. S., Poesch, M. S., Vinebrooke, R. D., Carli, C. M., & Taylor, M. K. (2019). Environmental DNA in lake sediment reveals biogeography of native genetic diversity. *Frontiers in Ecology and the Environment*, 17(6), 313-318. <https://doi.org/10.1002/fee.2073>.

Thomson-Laing, G., Parai, R., Kelly, L. T., Pochon, X., Newnham, R., Vandergoes, M. J., Howarth, J. D., & Wood, S. A. (2020). Development of droplet digital Polymerase Chain Reaction assays for the detection of long-finned (*Anguilla dieffenbachii*) and short-finned (*Anguilla australis*) eels in environmental samples. *PeerJ*, 9. <https://doi.org/10.7717/peerj.12157>

Naro-Maciel E, Ingala MR, Werner IE, Reid BN, Fitzgerald AM (2022) COI amplicon sequence data of environmental DNA collected from the Bronx River Estuary, New York City. *Metabarcoding and Metagenomics* 6: e80139. <https://doi.org/10.3897/mbmg.6.80139>