Neurospora DNA Extraction Protocol

Conidial Suspension

*Incubate Neurospora stab for at least 6 days

1. Add 10mL of Sterile H_2O to the desired stock culture
2. Vortex culture vigorously until conidia on glass and agar go into suspension
3. Strain resulting supernatant through sterile cheesecloth to remove agar and mycelia
4. Pour resulting conidial suspension into a centrifuge tube and centrifuge @ 3000 RPM for 10 minutes
5. Carefully pipette off the supernatant and discard
6. Resuspend conidia by bringing volume up to 250[microliters] with dH_2O and vortexing

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Qiagen Blood and Tissue Protocol

1. Add 180µL Buffer ATL
2. Add 20µL proteinase K at 56°C until completely lysed. Vortex occasionally.
3. Vortex for 15s. Add 200µL Buffer AL to the sample and mix thoroughly by vortexing. Then add 200µL ethanol (96%-100%), and mix again thoroughly by vortexing
4. Pipet the mixture from step 3 (including any precipitate) into the DNeasy Mini spin column placed in a 2ml collection tube (provided). Centrifuge at 8000 RPM for 1 minute. Discard flow-through and collection tube.
5. Place the DNeasy Mini spin column in a 2 ml collection tube (provided), add 500µL Buffer AWL1, and centrifuge for 1 minutes at 8000 RPM. Discard flow-through and collection tube.
6. Place the DNeasy Mini spin column in a new 2mL collection tube, add 500µL Buffer AWL 2, and centrifuge for 3 min at 14,000 RPM to dry the DNeasy membrane. Discard flow-through and collection tube.
7. Place the DNeasy Mini spin column in a clean 1.5mL or 2mL microcentrifuge tube (not provided), and pipet 200µL Buffer AE directly onto the DNeasy membrane. Incubate at room temperature for 1 min, and then centrifuge for 1 min @ 8000 RPM to elute.
8. Recommended: For maximum DNA yield, repeat step 7.