Supplementary protocol

- Supplied by a customer

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NucleoSpin® Tissue – purification of DNA from methicillin-resistant *Staphylococcus aureus* (MRSA) (Rev. 01)

This protocol is only a supplement to the kit's general user manual. Please refer to the kit manual for more detailed information regarding safety instructions, product-specific disclaimers, and especially preparations needed before starting the procedure. The latest version of the user manual is available at ***www.mn-net.com/usermanuals*** *or can be requested from our technical service (tech-bio@mn-net.com). Material safety data sheets (MSDS) can be downloaded from* ***www.mn-net.com/MSDS****.*

Additional equipment needed:

- Water bath(s) or heating block(s)
- Lysozyme (10 mg/mL) and lysostaphin (10 mg/mL)

Additional preparations before starting:

- Set incubators or water baths to 37 °C and 70 °C, respectively.

1 Prepare sample (swabs in Stuart Medium)

Vortex vigorously for **1 min** in order to remove all bacteria cells from the swab. Remove the swab and centrifuge the sample for **5 min** at **8,000 x *g***. Remove and discard supernatant.

2 Pre-lyse cells

Resuspend the pellet in **160 µL 50 mM EDTA (pH 8)** by pipetting up and down. For efficient lysis, add **20 µL** of **10 mg/mL lysozyme** and **20 µL** of **10 mg/mL lysostaphin**. Vortex vigorously and incubate for **30–60 min** at **37 °C**. Vortex occasionally during incubation or use a shaking incubator.

3 Lyse cells

Add **200 µL Buffer B3**, vortex vigorously and incubate at **70 °C** for **10 min**. Vortex briefly.

Proceed with step 4 of the standard protocol ('Adjust DNA binding conditions').

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