analytikjena
An Endress+Hauser Company

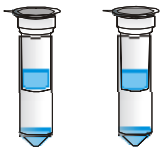
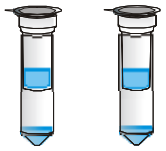
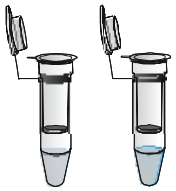
innuPREP DNA / RNA Mini Kit

Protocol 3: DNA and RNA extraction from bacterial cells

Recommended steps before starting

- Prepare Washing Solution HS and Washing Solution LS according to the instruction

1. Starting material	▪ Bacterial cells		▪ Max.: 1 x 10^9
2. Pellet cells			▪ 5,000 x g (~7,500 rpm); 5 min ▪ Discard supernatant
3. Lysis of cells			▪ Add 100 µl TE buffer ▪ Resuspend cell pellet ▪ Add Lysozyme (gram- 2 µl; gram+ 6 µl) ▪ Incubation (clear or viscous sol.) ▪ Add 450 µl Lysis Sol. RL, vortex ▪ Incubation: 3 min @ RT
4. Binding of gDNA New Receiver Tubes			▪ Centrifuge: max. speed;1 min ▪ Spin Filter D to Receiver Tube ▪ Add supernatant to Spin Filter D ▪ 10,000 x g (~12,000 rpm): 2 min ▪ Spin Filter D to Receiver Tube ▪ **Don't** discard the filtrate!
5. Binding of RNA Re-use Receiver Tube			▪ Add equal volume 70 % ethanol (approx. 600 µl) to filtrate ▪ Spin Filter R to Receiver Tube ▪ Add 650 µl filtrate to Spin Filter R ▪ 10,000 x g (~12,000 rpm): 1 min ▪ Load residual sample ▪ 10,000 x g (~12,000 rpm): 1 min ▪ Spin Filter R to Receiver Tube

7. Washing of Spin Filter D and R Re-use Receiver Tube		■ Add 500 µl Washing Solution HS ■ 10,000 x g (~12,000 rpm): 1 min ■ Add 700 µl Washing Solution LS ■ 10,000 x g (~12,000 rpm): 1 min
8. Remove Ethanol of Spin Filter D and R Re-use Receiver Tube		■ Discard filtrate ■ Spin Filter D to Receiver Tube ■ Spin Filter R to Receiver Tube ■ Centrifuge: max speed, 2 min
9. Elution of Spin Filter D and R		■ Spin Filter D to an Elution Tube ■ Add 100 µl Elution Buffer ■ Spin Filter R to an Elution Tube ■ Add 30-80 µl RNase-free Water ■ Incubation: 1 min @ RT ■ 6,000 x g (~8,000 rpm): 1 min

Order No.:	845-KS-2080010	10 reactions
	845-KS-2080050	50 reactions
	845-KS-2080250	250 reactions

This documentation describes the state at the time of publishing. It needs not necessarily agree with future versions. Subject to change!

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