Quick-Start Protocol November 2016

QIAamp® BiOstic® Bacteremia DNA Kit

The QIAamp BiOstic Bacteremia DNA Kit can be stored at room temperature (15–25°C) until the expiry date printed on the box label.

Further information

- Safety Data Sheets: **www.qiagen.com/safety**
- Technical assistance: **support.qiagen.com**

Notes before starting

- Warm Solution MBL at 55°C for 5–10 min to dissolve precipitates prior to each use.

1. Swirl the cultured blood to resuspend the bacteria and remove 1.8 ml with a syringe and needle or pipette and dispense into a 2 ml collection tube (provided).
2. Centrifuge at 13,000 x *g* for 2 min to pellet the bacteria and pipette to remove the supernatant and dispose in biohazard waste.
3. Add 450 µl of Solution MBL to the pellet and resuspend by pipetting. Transfer the lysate into a 2 ml PowerBead Tube Garnet 100 and close. Vortex for 10 s to mix and place in a 70°C heat block or water bath for 15 min.
4. Secure the PowerBead Tube horizontally using the Vortex Adapter tube holder for the vortex (cat. no. 13000-V1). Vortex at maximum speed for 10 min.
5. Centrifuge the PowerBead Tube to pellet debris at 10,000 x *g* for 1 min. Transfer the supernatant to a new 2 ml collection tube (provided).
6. Add 100 µl of Solution IRS and vortex to mix. Incubate for 5 min at room temperature.
7. Centrifuge at 10,000 x *g* for 1 min to pellet debris. Transfer the supernatant to a new 2 ml collection tube (provided).
   Note: Longer incubation in Solution IRS does not affect DNA yield or purity (Sample may be incubated up to 10 min in Solution IRS).

8. Add 1 ml of Solution BB. Pipette or pulse vortex to mix. Briefly centrifuge to collect any liquid from the top of the lid.
9. Load 600 µl of lysate onto a MB Spin Column and centrifuge at 10,000 x *g* for 1 min.
10. Discard the flow-through and place the MB Spin Column back into the 2 ml collection tube. Repeat this step until all the lysate has been loaded onto the Spin Column.
11. Transfer the MB Spin Column to a new 2 ml collection tube (provided) and wash by adding 500 µl of Solution CB. Centrifuge 10,000 x *g* for 1 min. Discard the flow-through and put the MB Spin Column back into the 2 ml collection tube.
12. Wash with another 500 µl of Solution CB and spin at 10,000 x *g* for 1 min. Discard the flow-through and place the MB Spin Column back into the 2 ml collection tube.
13. Centrifuge at 13,000 x *g* for 2 min to dry the MB Spin Column membrane.
14. Transfer the MB Spin Column to a new 2 ml collection tube (provided).
15. Elute by adding 50 µl of Solution EB directly in the center of the membrane. Allow the MB Spin Column to sit at room temperature for up to 5 min to maximize the elution.
    Note: Do not heat the elution buffer.
16. Centrifuge at 10,000 x *g* for 1 min.
17. Discard the MB Spin Column and cap the 2 ml collection tube containing the genomic DNA. The DNA is now ready for downstream applications.
    Note: We recommend storing DNA frozen (–20° to –80°C) as Solution EB does not contain EDTA.