

SureFast® Clostridium botulinum Screening PLUS DNA-extraction from honey based on DIN CEN ISO/TS 17919

Art. No. F5110

March 2021

1 Sample preparation

For the detection of Clostridium botulinum in honey the samples have to be prepared specially.
Please prepare the samples in duplicates.

1.1 Equipment and consumables

- 2 ml reaction tubes and 50 ml falcon tubes
- Centrifuge with 2 ml and 50 ml inserts
- Water bath with shaker
- PCR grade water with 1 % Tween 80, heat to 50 °C ± 1 °C
- SureFast® PREP Aqua (F1023)

1.2 Procedure

- Warm up the honey in a water bath with integrated shaker for 30 min at 50 °C ± 1 °C and 700 rpm
- Weigh in 2 x 12.5 g of honey sample in 50 ml Falcon-tubes
- Add 25 ml of preheated PCR grade water with 1 % Tween 80 (50 °C ± 1 °C)
Please prepare and analyze the PCR grade water with 1 % Tween 80 as an additional extraction control („EC Medium“)
- Centrifuge samples at 5.000 rpm for 30 min
- For the enrichment store the sediment in a refrigerator
- Filtrate the supernatant (Filter 0.45 µm) as described in SureFast® PREP Aqua
- When the filter is blocked use a new one
- In the following steps use all filters

1.3 Enrichment

- Preheat TPGY medium up to 65 °C ± 1 °C
- Put all filters from one sample in a 50 ml falcon tube
- Add 10 ml TPGY medium
- The filters has to be covered with TPGY medium
- Add 10 ml TPGY medium to the sediment (stored in the refrigerator)
- Incubate the samples for 10 min at 65 °C ± 1 °C in a water bath
- Then the enrichment takes place