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Host DNA Depletion Kit

Product Number: DNK69

Shipping and Storage

-20℃

Components

Component	DNK69 50 preps
Buffer GB2	2×20ml
Benzonase (250U/μL)	50μL
Proteinase K	1×1.25mL

Description

This kit is suitable for differential lysis of human derived cells and removal of human derived nucleic acids from biological liquid samples (bronchoalveolar lavage fluid, serum plasma, liquefied liquid, pleural and peritoneal fluid, cerebrospinal fluid, swabs, etc.). BufferGB2 in the reagent kit selectively lyses host cells, releasing host nucleic acids that are rapidly degraded by nucleases, while bacteria, fungi, and other microorganisms experience almost no loss during this process. This product can be used in conjunction with silica gel membrane centrifuge column and magnetic bead nucleic acid extraction kits: while significantly reducing the host background, it specifically enriches microbial nucleic acids in the sample, greatly improving the sensitivity and specificity of PCR or real-time PCR analysis of pathogens, greatly enhancing the accuracy and sensitivity of mNGS detection, and has good application prospects.

Preparation and important precautions before the experiment

1. Thaw Buffer GB2 at room temperature or 2-8℃ before use and mix thoroughly. After thawing, BufferGB2 can be stored at 2-8℃ for 1-2 weeks without affecting its activity, and long-term storage should be at -20℃. To ensure optimal performance, freeze-thaw cycles should not exceed 3 times. If less than one bottle of BufferGB2 is required for extraction at a time, please ensure that it is used under sterile conditions such as an ultra clean workbench, and avoid contamination and growth of microorganisms in the remaining buffer solution.
2. To avoid erroneous results caused by pollution, please keep the work area clean and wear protective clothing, and set up control samples for quality control reasonably. Take appropriate measures to handle sample materials and reduce the risk of cross contamination. Immediately cover the bottle cap after using the reagent to prevent contamination.

Protocol

1. Take the pre processed sample and centrifuge it at room temperature at 10000rpm for 5-10 minutes. Be careful to discard the supernatant

   Note: Do not disturb the sedimentation of lower level cells to avoid sample loss.
2. Add 500μL of Buffer GB2, vortex and mix well, incubate at room temperature and 600rpm for 10 minutes.
3. Centrifuge at 12000rpm for 2 minutes, carefully remove the supernatant

   Note: Do not disturb the bacterial sediment when removing the supernatant to avoid sample loss.
4. Add 200μL BufferGB2 and 1μL Enzonase to the precipitate, incubate at 37℃ and 600rpm for 20 minutes. Add 20μL Proteinase K, vortex well, and incubate at 56 ℃ for 10 minutes. Instantaneous centrifugation after reaction completion
5. Immediately carry out subsequent microbial nucleic acid extraction to avoid affecting the extraction yield.