

Ready Reference for Yeast and Mold PCR Assay

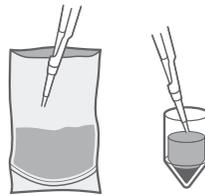
1. Homogenize sample in 1:10 dilution according to the food type.



2. Determine sample volume to be tested.

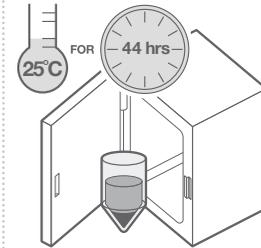
(See User Guide or table on back of this reference card.)

3. Transfer sample to disrupter tube.

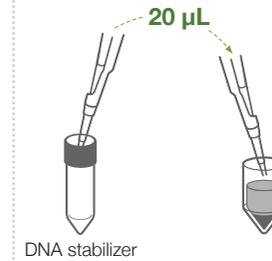


Pooled sample protocol requires triplicate disrupter tubes.

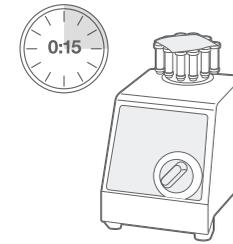
4. Incubate disrupter tubes.



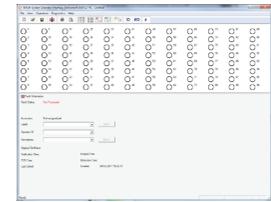
5. Add DNA stabilizer to disrupter tubes.



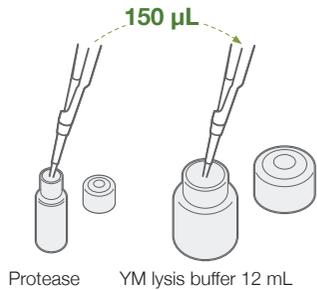
6. Agitate in disrupter device.



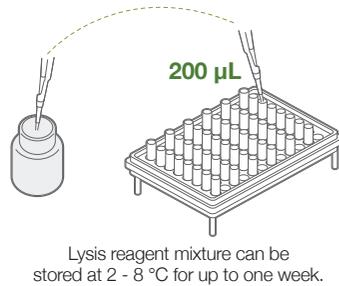
7. Create a rack file.



8. Add protease to YM lysis buffer.

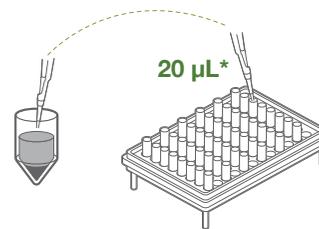


9. Transfer lysis reagent made in step 8 to cluster tubes.



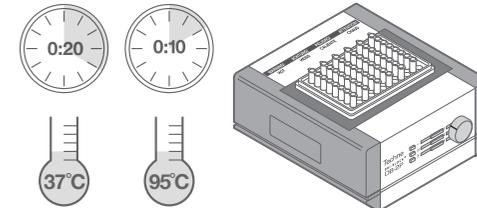
Lysis reagent mixture can be stored at 2 - 8 °C for up to one week.

10. Transfer disrupted samples to cluster tubes.

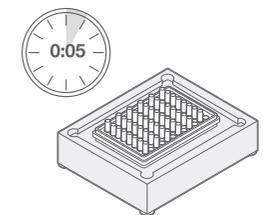


*Pooled sample protocol requires pooled volumes from disrupter tubes into 1 cluster tube.

11. Heat cluster tubes.

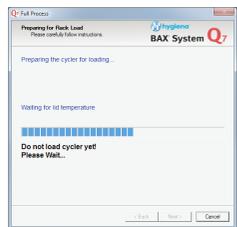


12. Cool cluster tubes in cooling block.

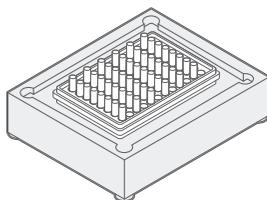


* Steps 11 and 12 can also be performed using the Hygiena[™] Automated Thermal Block. See the Automated Thermal Block User Guide for details and instructions.

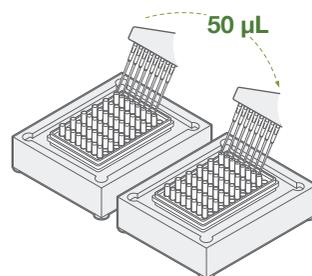
13. Initialize cycler.



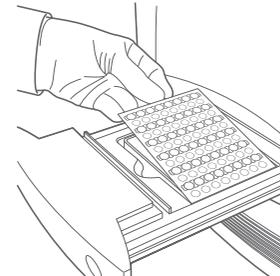
14. Arrange PCR tubes in cooling block.



15. Hydrate PCR tablets with 50 µl lysate from step 12.

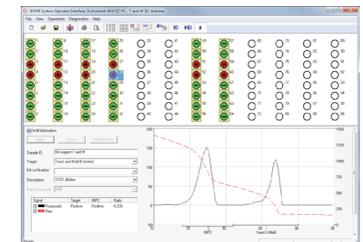


16. Place tubes in cycler and run program.



11. Unload samples and review results on screen. See User Guide for details.

- Negative
- Positive
- Indeterminate
- Signal error



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Pooled Sample Protocol

This ultra-sensitive protocol uses pooled samples from three disrupter tube enrichment replicates for action levels of 10-50 cfu/g.

If your action level is:	Then transfer this volume of homogenate to 3 disrupter tubes:	And pool these volumes of disrupted sample for testing:
10 cfu/g	400 µL	7 µL from 3 replicates
20 cfu/g	200 µL	7 µL from 3 replicates
50 cfu/g	80 µL	7 µL from 3 replicates

Non-Pooled Sample Protocol

This protocol for yeast and mold testing can be used without pooling for action levels of 25 cfu/g or above.

If your action level is:	Then use this volume of homogenate:
25 cfu/g	400 µL
50 cfu/g	200 µL
100 cfu/g	100 µL
500 cfu/g	20 µL
1000 cfu/g	10 µL