SeCore™ Workflow Quick Reference Card-CE-IVD

**Amplification**

1. Add DNA (15–30 ng/μL) to the bottom of each tube/well in the amount indicated to the right.
2. Create a mastermix, for N+1 samples, of Amp Mix and Taq using the volumes indicated to the right. Pulse vortex 2-3 times.
3. Add mastermix to the wells containing DNA [20 μL for Class I reactions and 23 μL for Class II reactions].
4. Cover and centrifuge briefly. Place plate in thermal cycler.

**ExoSAP-IT® Reagent Cleanup**

5. Remove 5 μL of PCR product and combine with loading dye. Load onto a 2% agarose gel to check for amplification.
6. Add 4 μL of ExoSAP-IT reagent to the bottom of each well. Centrifuge ~5 seconds.

**Sequencing Reactions**

8. Add 40 μL of ultra pure water to Class II reactions only. Vortex and centrifuge briefly.
9. Add 2 μL of ExoSAP-IT reagent-treated amplicon to a 96-well optical plate.
10. Add 8 μL of the appropriate sequencing primer to these same wells. Vortex and centrifuge briefly. Place plate in thermal cycler.

**Ethanol Precipitation**

11. Add 2 μL of PPT buffer to each well. Centrifuge briefly.
12. Add 40 μL of 100% (absolute) ethanol to each well. Vortex for 1 min.
13. Centrifuge for 30 min at 2,000 x g.
14. Invert on a paper towel and centrifuge inverted for 10–60 seconds at 500 x g.
15. Add 100 μL of 70%–80% ethanol to each well. DO NOT vortex.
16. Centrifuge for 5 min at 2,000 x g.
17. Invert on paper towel and centrifuge inverted for 1 min at 500 x g.
18. Add 15 μL of Hi-Di Formamide to each pellet.
19. Denature the samples at 95°C in a thermal cycler for 2 min.

**Find out more at onelambda.com**

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**Instrument Parameters**

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<th>Instrument</th>
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