

Xploregen Discoveries Pvt. Ltd.

SAMPLE VOLUME

<u>Clear water sample</u> - Syringe filter 50-200ml of the sample

Note: For density turbid sample take 50ml while for transparent sample take upto 200ml or more. Following the filtration step, open the filter, remove the membrane, cut the membrane into pieces, and add the pieces to the beaded vial containing the 1400 µl of XWA 1

IMPORTANT NOTES

- 1. Wear gloves and lab coat when handling the buffers
- 2. Add the required volume of ethanol (96-100%) to XWA 4 before use.
- 3. Vortex the beaded vial for 2-3 sec
- 4. Perform all the centrifugation steps at room temperature.
- Gently shake the reagents before using.

KIT CONTENTS (50/100/250)

- 1. XWA1-70ml/140ml/350ml
- 2. XWA2-10ml/20ml/50ml
- 3. XWA3-30ml/60ml/150ml
- 4. XWA4-30ml/55ml/130ml
- 5. XWA5-3ml/6ml/15ml

WATER DNA EXTRACTION PROTOCOL

- 1. Take 1 ml of Water (turbid) in a 1.5 ml sterile vial.
- 2. Centrifuge the tube at 10,000 rpm for 10 minutes at room temperature (RT) and discard the supernatant.
- 3. Add 1400 µl of **XWA 1** and resuspend the pellet completely by pipette mixing.
- 4. Add the complete amount of resuspended solution to the beaded vial.
- 5. Horizontal vortex the vial at maximum speed for 15 minutes.
- 6. Centrifuge the tube at 10,000 rpm for 2 minutes at room temperature (RT)
- 7. Transfer 700 µl of supernatant to a sterile 2 ml vial.
- 8. Add 200 µl of XWA 2 solution and vortex for 5 seconds
- 9. Centrifuge at 10,000 rpm for 2 minutes.
- 10. Transfer 750 µl Supernatant to a clean sterile 2ml vial.
- 11. Add 600 µl of **XWA 3** solution to the supernatant and vortex for 5 seconds.
- 12. Transfer 700 μl of Lysate to the spin column (Do not discard the residual lysate) and Centrifuge at 10,000 rpm for 2 minutes and discard the flow through.
- 13. Repeat the above (10&11) steps to collect all the Lysate (Ensure the entire lysate is processed via the spin column)
- 14. Add 500 µl of **XWA 4** to the spin column and Centrifuge at 10,000 rpm for 2 minutes and discard the flowthrough.
- 15. Centrifuge the empty spin column for 5 minutes at 10,000 rpm.
- 16. Place the spin column into a sterile 1.5 ml vial.
- 17. Add 30 µl of **XWA 5** (Elution 1) to the center area of spin column and centrifuge for 5 minutes at 10,000 rpm.
- 18. Place the spin column to a new sterile 1.5ml vial.
- 19. Add 25 ul of **XWA 5** (Elution 2) to the center area of spin column and centrifuge for 5 minutes at 10,000 rpm.
- 20. Discard the spin column and store both elution tubes for further processing.

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