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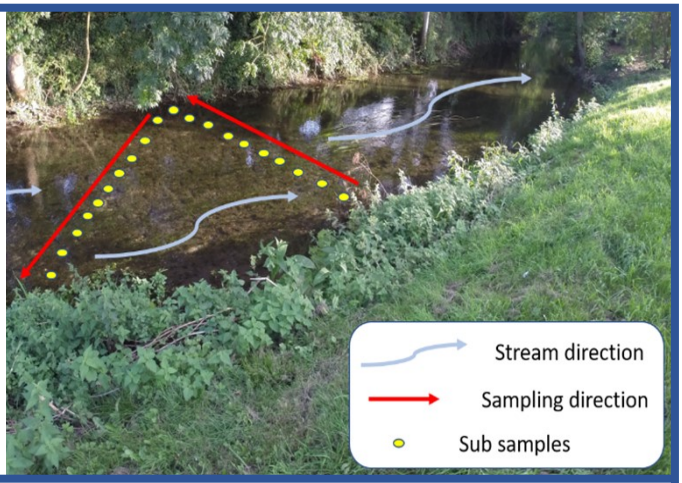
Detailed Filtration Sample Collection Guidance

1. Identify where 20 sub-samples will be taken from the river or pond perimeter.

The location of these should be spaced as evenly as possible around the site. In ponds, samples should be taken from locations around the entire pond perimeter, where accessibility permits. In rivers, samples should be taken against the flow of the stream, working upstream in a diagonal pattern where possible to ensure that any disturbed sediment is not collected, should it be necessary for the collector to enter the watercourse.



2. Wearing gloves, open the sterile Whirl-Pak bag and collect 20 ladles of water from around the site.

The water sample should be taken from the middle of the water column. Where possible, avoid any disruption of sediment as this can both clog the filter quicker and introduce ancient DNA into the sample. In larger sites it may be necessary to use a telescopic pole.

Once collected close the bag securely and shake to mix the water sample.

3.

Using the large syringe, take 50ml of sample from the Whirl-Pak bag.

Attach the syringe using a half twist action to the filter unit. The syringe will only fit to one end of the filter unit. Note, twisting too far can damage the luer lock connection on the filter.

Apply pressure to the syringe until all liquid has passed into and through the filter.

Remove the syringe from the filter and repeat the process until:

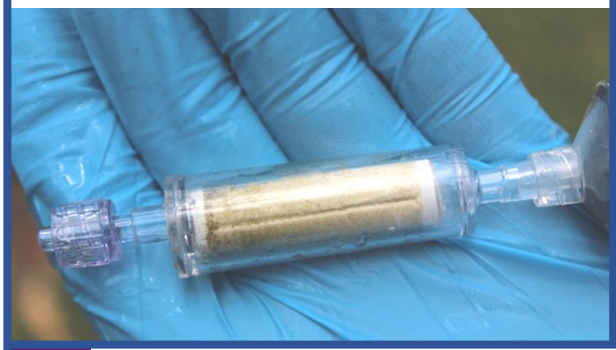
A) you have filtered 500ml OR

B) The filter has become blocked and cannot filter any more

Record the volume of liquid which has been filtered on the sample collection form.

Instructions continue overleaf

4. Empty the syringe and fill with air, re-attach to the filter and push air through the filter unit until it is completely free of water.



5.

Screw one red cap tightly on to the thick end of the filter unit.

Place the filter unit to one side.

6. **Using the small blue syringe, collect up 2ml of preservative solution from the preservative tube.**

An excess of preservative solution is provided.

It is important to add preservative solution into the filter unit to prevent sample degradation during transport to the laboratory.

7. **Attach the syringe to the open end of the filter unit.**

Apply light pressure until all 2ml of preservative solution is within the filter casing.

Repeat process if necessary until internal casing is filled by preservative solution.

8. **Finally, screw the second red cap on to the filter inlet.**

Ensure that both caps are secured tightly to avoid leakage of preservative solution during transport to the laboratory.

Place the sample into the 50ml tube provided and return to laboratory.

Samples can be stored at room temperature for up to 3 weeks. Longer if chilled.