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We use this protocol and it's working

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Field sampling for drinking water

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ABSTRACT

The method we used for sampling drinking water for households when detecting *S.* Typhi and Paratyphi in Nepal

GUIDELINES

Sterility during water sample collection
1. Sterility of collected water sample: The primary risk of sample cross-contamination comes
from either the field assistant or water/DNA from another household.
a. Field assistant: It is important that the field assistant, whom comes into contact with
water at multiple households, uses proper technique and does not contaminate the water being collected by touching or handling the opening or inside of the bag.
b. Collection instrument: while the inside of each WhirlPak is sterile, the outside is not.
Thus, a Whirlpak cannot be directly dipped into a water tank to collect water since it is not sterile, has been handled by the field assistant and may have come into contact with another household's water during transport. Items owned and used by the household are an exception. Since water at the household level is routinely transferred via multiple non-sterile methods (pipes, valves, jugs, cups, etc) prior to consumption, it is ok, for water to be in contact with these devices prior to collection.
2. Contamination of household water: It is similarly important that potential pathogens are not
introduced into a water supply when we collect from there.

Definitions:

- Origination of water: Defined as the principal, original source of water. For water that is
  processed or distributed (i.e. municipal or private water company) then the initial water
  treatment and distribution plant is the origination point. For unprocessed water (i.e.
  collected or distributed directly from ground water or surface water for

consumption),
then the source of water prior to distribution or collection is the origination (i.e. river or
groundwater).

- Freshness: Refers to time since water is delivered from the origination source. For
instance, the freshest possible water would be immediately collected from a pipe that
has actively running water from a municipal water source. Water that has been stored
and has been stagnant/still for an amount of time, freshness decreases. Thus, water
collected from a running pipe and stored for 24 hours would be less fresh then running
pipe water but fresher then water collected and stored 1 week prior.
- End-user treatment: Any treatment of water done at the household level prior to use or
consumption, including but not limited to filtration, boiling or chemical treatment such as
chlorination.

MATERIALS

Materials list
1.Gloves
Whirl-Pak bag (Nasco product Number: B01027) or sterile bottle
2.Barcode Labels (blue, printed in duplicate)
3. Bag locking pipe closures (Nasco product number: B01595)
4. Ice packs
5. Insulated bag or cooler box
6. Empty bottle sleeves—plastic bottle with the top cut off (for keeping bags/bottles upright during
transport)
7. 2 sterile water pumps (for difficult to reach water collection)

1 All water will be collected at the point closest to the origination of that water supply prior to end-user treatment to minimize the time for S. Typhi or Paratyphi DNA to degrade in
water, if present. If the only option is to sample from water that has been stored, choose the freshest (most recently stored) source.

Sample Collection

2 Pre-label all Whirl-Pak bags with a fill line representing 1 litre of water
a. Fill a bag with exactly 1 litre of water in the lab. Mark the outside of the bag with a

line where the water reaches. Copy this line to the rest of the Whirl-Pak bags.

3 Place two duplicate random number stickers on the Whirl-Pak bag.

4 Put on gloves and ipen the bag, taking care not to touch the inside of the bag

5 Fill the bag with 1 liter or water

5.1 If necessary, ask the household member to turn on the water spigot so that you do not contaminate your gloves.

5.2 If the household uses water from a communal spigot, collect from this point
i. If a hose is connected to a communal spigot, remove it to collect directly from the tap

5.3 If the community spigot is not functioning/the water is not running, sample water from the house or building collection tank, using the sterile hand pump

5.4 If the storage tank in inaccessible, you can sample from stored water in a jug
i. Choose the freshest source of stored water (the one that was stored the most recently)
ii.Avoid using a cup or container to transfer water from the jug If the container is too heavy to lift and pour into the Whirl-Pak bag, you can request to use the same cup or device the household uses to transfer water.

6 Close the bag with the pipe-locking device

7 Remove gloves

8 Scan the sample barcode into RedCap

9 If the code does not scan, manually enter it

10 Take a picture of the sample in the RedCap app, making sure that you can see the barcode number

Sample transport

11 Place the full bag Whirl-Pak in an upright plastic bottle inside the cooler bag/backpack

11.1 Unused Whirl-Pak bags should be kept in a separate compartment from water samples

12 Minimize opening and closing the cooler to maintain temperature control.

13 If maintaining temperature control is a challenge, consider placing temperature loggers inside each cooler bag.