

## FROZEN FINGER PROTOCOL

### SUPPLY LIST

#### For freezing process

1. Finger/rod with crossbar handle for extraction
2. denatured alcohol (can get 4+ cores per 3 gallon container, plus some for cleaning)
3. ~5 lbs dry ice per core; 5 lbs to remain in cooler to keep samples frozen
4. Cooler
5. Mallet
6. Large serving spoon
7. Trowel
8. Funnel to fit in top of finger
9. Face protection
10. Insulated gloves
11. Meat grinder (optional)
12. Pipe wrench (if using smallest diameter corer, not recommended)
13. Pokey stick (if using smallest diameter corer, not recommended)

#### For subsampling

1. Piezo electric propane or map gas torch
2. Kim whipes, paper towels, or clean shop towels
3. Tarp and parachute cord for shade if sampling date is hot or rainy
4. Non-serated chef's knife
5. Ziplock bags (pre-weighed and pre-labelled)
6. Felt pen
7. Measuring tape
8. Camera
9. Nitrile gloves (~20) and two pairs of leather, or insulated gloves
10. Aluminum foil
11. Steel rod or trowel for mixing EtOH/dry ice slurry
12. Mallet

### INSTRUCTIONS:

1. Pre-label ziplock bags. See below for labeling method.
2. It is best to do this process as early in the morning as possible to avoid the mid-day heat on hot summer days. Set up tarp if necessary.
3. The freeze corer does not work well with non-saturated material. Therefore, if the water table is below the surface it is better to sample the 0-10 cm increment by hand with scissors. Be sure to clean (flame and alcohol, see below for more info) the scissors and to wear gloves when sampling.
4. Break dry ice into small pieces by pounding it with the end of a mallet (up and down movement). If dry ice is from Fred Meyer, this is best done while it is still in its bag. If dry ice is from Arctic Fire & Safety crush it in one of the 5 gal buckets.
5. Powderize dry ice: *Note: this method did not work with the fresh dry ice purchased from Arctic Fire and Safety. It appears that the meat grinder only works well with older dry ice from Fred Meyers. Also, this process does not need to be done with the 2" diameter corer but probably should be attempted with the 1" diameter corer.* Place pieces of dry ice into meat grinder and grind to a powder. Placing the grinder over the side of a cooler so that the 'snow' falls into the cooler works well.

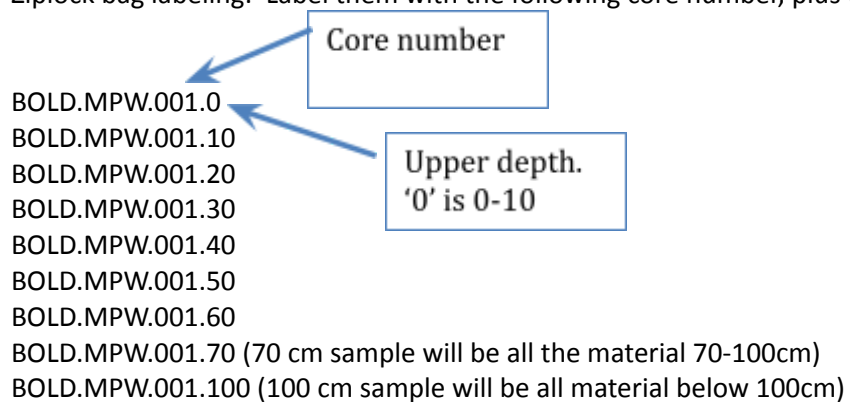
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6. Prepare work area by placing aluminum foil on boardwalk and having the measuring tape, knife, camera, propane torch (for cleaning) and pre weighted pre labeled plastic bags in place.
7. Flame the rod with torch and wipe down with towel soaked in alcohol to fast cool. Insert rod into peat until it is at least 70 cm deep. Be aware that the flame treatment removes contaminating DNA from one core to the next. Alcohol will sterilize a core, but actually preserve contaminating DNA.
8. Combine denatured alcohol and dry ice and mix. Add dry ice again until it creates a thin slush (like a slushy). While wearing eye/face protection, carefully spoon the slurry into the tube using a funnel (this is especially important if using the 1" diameter corer). Use a stick or core rod to poke the slurry through the funnel if it gets stuck. Fill corer until you can see frost along the exposed surface of the corer. Check the surface of the soil for a rind of frozen soil/water. Note: For the smaller diameter corer you will need ~300 ml of slurry to fill the finger to 150 cm depth, but much of it can be lost through bubbling, so you will need to make ~800 ml of slurry. For the 2" diameter corer much less of the slurry is lost (usually fine with slurry made with 5 lbs of dry ice).
9. Using the crossbar handle, break the finger loose by turning it several times and then extract the core from the peat. It is easiest to extract if you turn a bit as you're lifting up.
10. Carefully pour the slurry back into your bucket. It can help to have someone raise the bucket up to the corer opening, then lower the corer and the bucket together.
11. Working QUICKLY while wearing gloves lay the finger on aluminum foil on the boardwalk alongside measuring tape (IN THE SHADE!). Take photos down the length of the frozen sleeve so that the tape can be seen in each picture (3-4 photos).
12. Flame the knife and wipe with alcohol. Place the knife at the location you would like to cut and bang on it with the mallet. Even if you're taking 10 cm increments it is easier to cut portions off in 2-3 cm increments. It works well to have one person turning the core and making sure the knife surface stays clean. The other person keeps one hand clean and only uses it to touch the sample, using the other hand to hold the ziplock bag. Do not pick up material that falls onto a muddy surface.
  - a. If you have three people you can have one person can hold the handle of the rod slightly in the air. The second person stands on bottom of the rod for stability (this bottom spot won't be used). The third person sits perpendicular to the core and marks the frozen sleeve every 10 cm down to 70 cm. Assistant at head or foot of finger holds open the ziplock bag while person sitting perpendicular to the core taps the sample so it falls into the bag.
13. Place bag immediately into cooler with dry ice.
14. Between depth segments, wipe off knife and re-clean it (flame & alcohol). Change gloves.
15. Write down the photo numbers that were taken for each core in your field book.
16. Measure water table depth at site and record.

If Fred's doesn't have dry ice, you will need to go to Arctic Fire and Safety (702 30th Ave, Fairbanks, AK 99701). YOU WILL NEED TO CALL A DAY AHEAD (800 260 3463) to place an order for the following morning, since they make the dry ice on site. They may require a credit card when you call in.

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Ziplock bag labeling: Label them with the following core number, plus the date.



Do same sampling scheme for the next two cores: BOLD.MPW.002.0 and BOLD.MPW.003.0