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Sampling Whale Bones for Genetic Analysis

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AND

McLeod BA, Brown MW, Moore MJ, Stevens W, Barkham SH, Markham M, White BN (2008) Bowhead whales, and not right whales, were the primary target of 16th- to 17th-century Basque whalers in the western North Atlantic. *Arctic* **61**: 61-75.

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We use this protocol and it's working

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Abstract

This protocol describes how to collect samples from whale bones. This protocol is most often used for historical (ancient) samples of bone and has been optimized for downstream ancient DNA analyses. The most important issue with genetic analysis of historical samples is contamination. Therefore, to minimize the risk of this, we throw away most of the materials between sampling events (gloves, paper towels, etc.), and wash the re-usable materials (drill bits) in bleach between samples.

Guidelines

We clean all of our work spaces and pipettes with a 10% bleach solution. This has been shown to effectively clean cells off of laboratory equipment (Merritt et al. 2000), as well as to degrade DNA to a degree where it is no longer available to act as a template in PCR (Prince & Andrus 1992; Kemp & Smith 2005).

Materials

	Supplies
	Weigh boat (one per sample)
	Small glass beakers or 2 square tupperware container
	Sharpies
	Paper towel
	Notecards
	Kimwipes
	Disposable Nitrile gloves (one per sample)
	Cordless drill
	Clean filtered water
	Camera
	Bleach
	4-6 titanium drill bits
	15 ml falcon tubes

Safety warnings

Use caution when using drill, as bone may sometimes because slippery due to oils.

Set up work space

1 Clean work area with 10% bleach solution including the outside of the drill. Lay down paper towels to cover the area where you will be drilling.

2 Fill one glass beaker or tupperware container about half full with clean, filtered water.

3 Fill the other beaker or tupperware container about half full with a 10% bleach solution.

Sampling Bones

4 Move the current bone you are sampling onto the designated work area, ensure the area is covered with paper towel.

5 Make a label for the bone using a notecard and sharpie and lay the label next to the bone. This label should include the ID of the sample, the date, and any other important information. Take a picture of the bone and label together.

6 Label a 15 ml falcon tube with the bone label.

7 Place a clean weigh boat on the paper towels.

8 Put on a clean pair of gloves over previous gloves and put a new clean drill bit into the drill.

9 Drill ~3 “starter holes” into the bone. These are shallow holes used to remove the surface of the bone which may be contaminated. You should allow these shavings to fall onto the paper towel as they are unwanted. Turn the bone upside down and shake to make sure all the shavings are gone from the bone.

10 Remove the drill bit and place it into the beaker/container with the bleach solution.

11 Put a new clean drill bit into the drill and position the weigh boat under the bone so that all shavings are collected.

12 Drill into the bone (where you made the starter holes), swirling the drill bit to try and maximize the amount of shavings obtained. Collect the shavings in the weigh boat.

13 Carefully transfer the shavings into the labelled 15 ml tube. A scoopula cleaned in 10% bleach solution may be used here to aid in the transfer. Scoopulas must be cleaned between samples with 10% bleach solution.

14 Place the drill bit into the 10% bleach solution and move any previous drill bits in the bleach solution into the water solution.

14.1 **Note**: The drill bits should go through a rotation of use ⇒ 10% bleach ⇒ water ⇒ use . . . and so on. After a drill bit is used it should go into the bleach solution for at least a few minutes (e.g., while the next bone is being sampled). Once it has been in the solution for long enough, it can be moved to the water to rinse. From the water, they can be blotted dry and used again. This is why it is necessary to have 2-3 pairs of bits as it ensures that each pair has enough time to soak in the bleach and then be rinsed in the water prior to being used again. Bleach is excellent at denaturing DNA as well as removing cells from utensils. If many bones are being sampled, the bleach and water solutions should be replaced every 10 samples as the solutions will become too dirty.

15 Throw away outer gloves, weigh boat, and paper towels. Set up area again for next bone and put on new clean gloves. Repeat steps above for as needed for the number of samples you are doing.

Protocol references

Kemp BM, Smith DG (2005) Use of bleach to eliminate contaminating DNA from the surface of bones and teeth. Forensic Science International 154: 53-61.

Merritt K, Hitchens VM, Brown SA (2000) Safety and cleaning of medical materials and devices. Journal of Biomedical Materials Research 53: 131-136.

Prince AM, Andrus L (1992) PCR: How to kill unwanted DNA. BioTechniques 12: 358-360.