

DNA profiling by SnotBot® sampling from the spout of a whale



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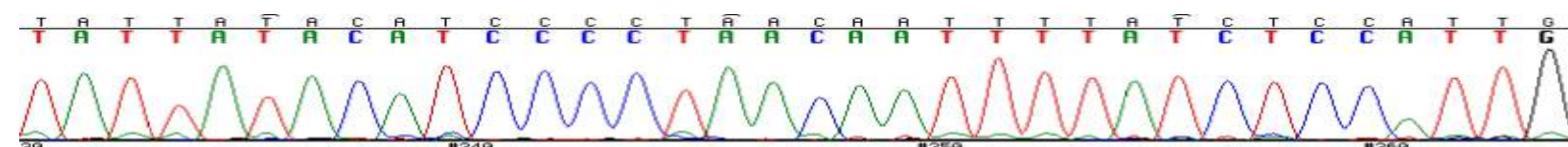
Research objectives

Sampling the spout of a whale using an aerial drone or SnotBot®, offers a non-invasive alternative to biopsy sampling. However, most non-invasive sampling techniques provide low quantity and quality of DNA, reducing the success of a DNA profile. Here, we report on methods to quantify the quality and concentration of DNA extracted from the spout of the whale using SnotBot® between 2016 and 2018.



Methods

We extracted DNA using either a phenol chloroform or Qiagen column method from biological samples collected from the spout of the whale and assessed the concentration of DNA within the extract using digital droplet PCR (ddPCR). The estimated copy number of target DNA as determined by the ddPCR was compared to the 'success' of the sample as determined through conventional PCR and sequencing of the mtDNA control region. For samples that yielded high-quality sequence, we assigned mtDNA control region haplotypes based on comparisons to sequences on GenBank.



Results

- Success of amplification and mtDNA sequencing improved from 58% in 2016 to 83% in 2018 (see Table 1). Success with amplification of other markers (e.g. sex and microsatellite loci) was more variable but also improved with time. The mtDNA haplotypes of SnotBot® samples were compared to worldwide databases and identified matches to blue whales reported in the North Pacific and humpback whales in the Gulf of Alaska and the Southern Hemisphere. In addition to confirming presence of haplotypes in sampled regions, we also identified previously unreported haplotypes.

Table 1. Total number of SnotBot samples collected (n) in Southeast Alaska (SEAK), Baja California, Gulf of Maine and Gabon. For each expedition, species detected is listed (Humpback whale, Mno; Blue whale, Bmu, killer whale, Oor; fin whale, Bph) as well as average copies/μl of DNA per sample, number and percentage of samples that provided information on sex and mitochondrial control region (mtDNA).

Year	Location	Species	n	copies/μl of DNA	sex	Percentage Success (sex)	mtDNA	Percentage Success (mtDNA)
2016	SEAK	Mno	31	3.22	13	41.94%	18	58.06%
2017	Baja	Bmu	9	2.55	2	22.22%	6	66.67%
2017	SEAK	Mno (Oor)	16	0.99	5	31.25%	6	37.50%
2018	Baja	Bmu	12	2.29	3	23.08%	7	58.33%
2018	Maine	Mno (Bph)	10	7.97	7	70.00%	10	100.00%
2018	Gabon	Mno	23	12.41	22	91.67%	20	83.33%
			Total	Total	Total	Average	Total	Average
			101	5.00	53	50.90%	66	61.38%

Funding thanks to:



Figure 1. Logistic regression model comparing ddPCR concentration to mtDNA sequencing success.

- Using a logistic regression, we estimated the concentration of DNA providing a 50% likelihood of amplification and sequencing of mtDNA to be 0.6 copies/μl.
- The lowest concentration of DNA with a positive result was similar to the lower threshold for successful barcoding with environmental DNA collected from seawater in the path of whales.



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- We report successful extraction and quantification of DNA from samples collected by SnotBot®
- With improved field methods and laboratory extraction techniques, we show an increase in the likelihood of obtaining a DNA profile.

