



CETUS PROJECT

Environmental DNA as a cetacean monitoring tool in the Northern Coast of Continental Portugal



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Background

- Increasing anthropogenic threats allied with cetacean research constraints lead to the urgency of finding new sampling methods to obtain data;
- That is where eDNA comes in, currently being an emerging complementary sampling technique for marine biodiversity monitoring programs;
- END GOAL:** Develop and optimize a methodology that allow us to successfully detect and identify cetacean species present in the Northern Portuguese Coast, from water samples without associated sightings.

Methods

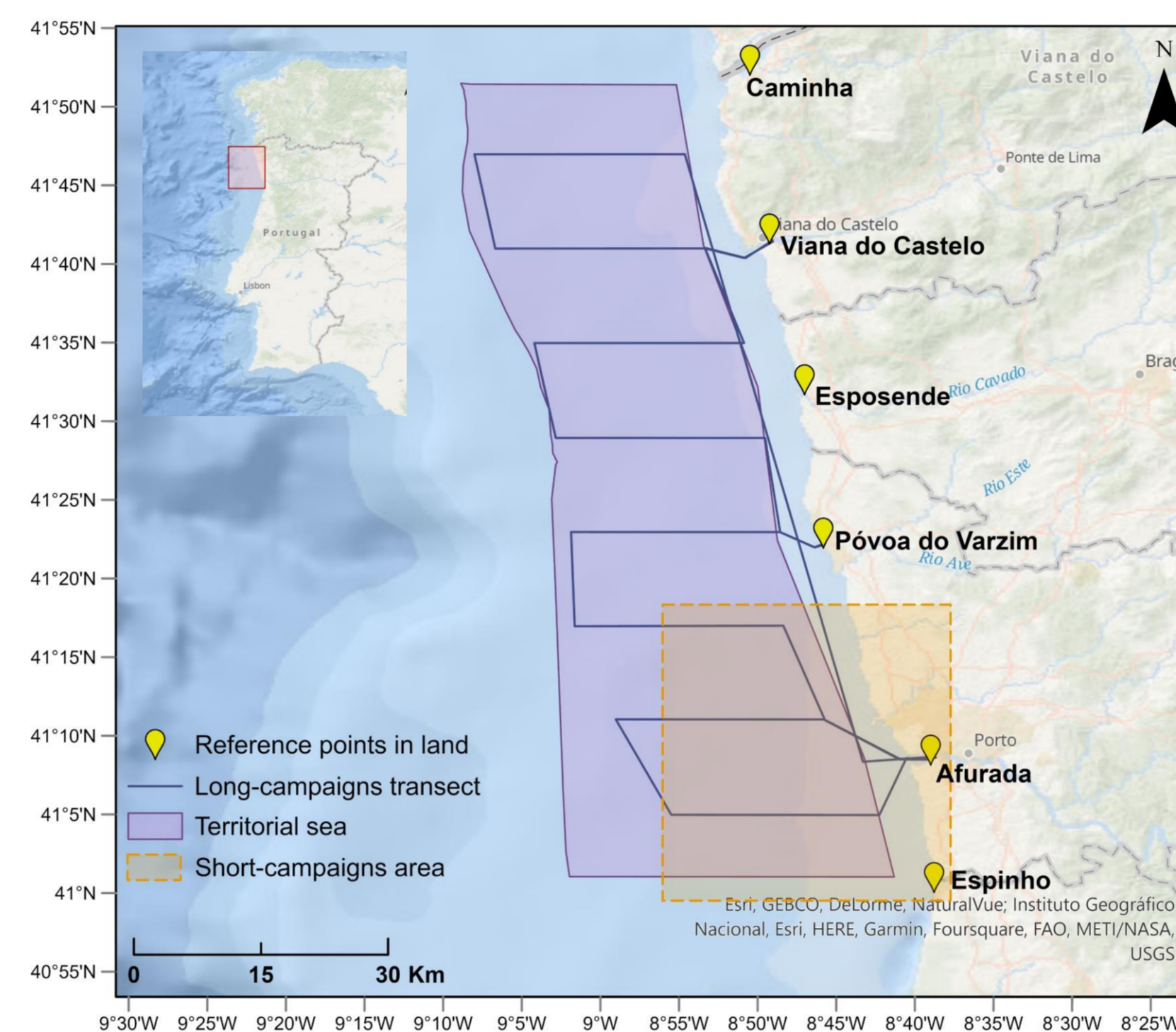


Fig.1- Study area in dedicated at-sea surveys in the Northern Portuguese Coast, from Espinho to Caminha up to 10 nautical miles from coast

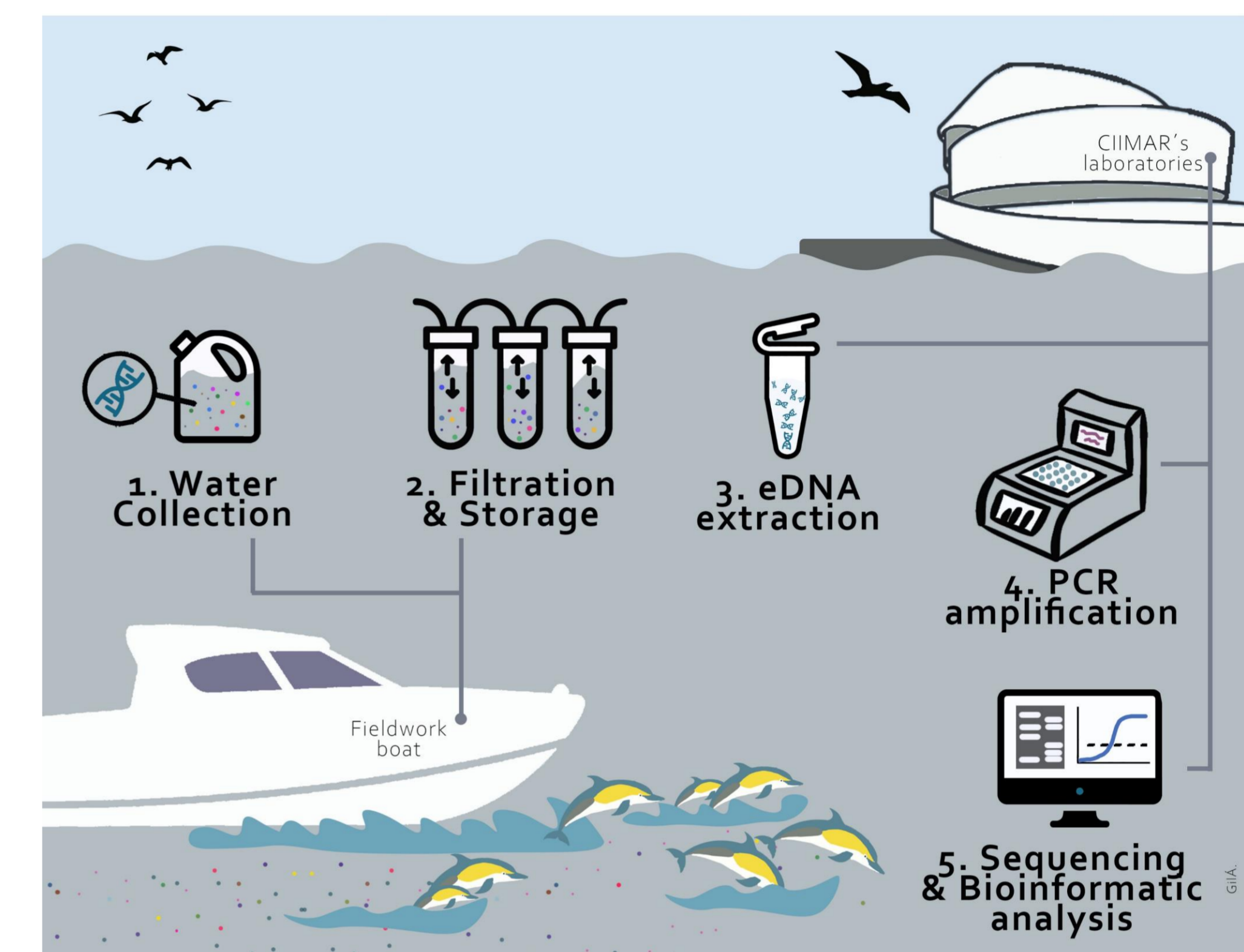


Fig.2- eDNA methodology steps applied for marine mammals monitoring

Results

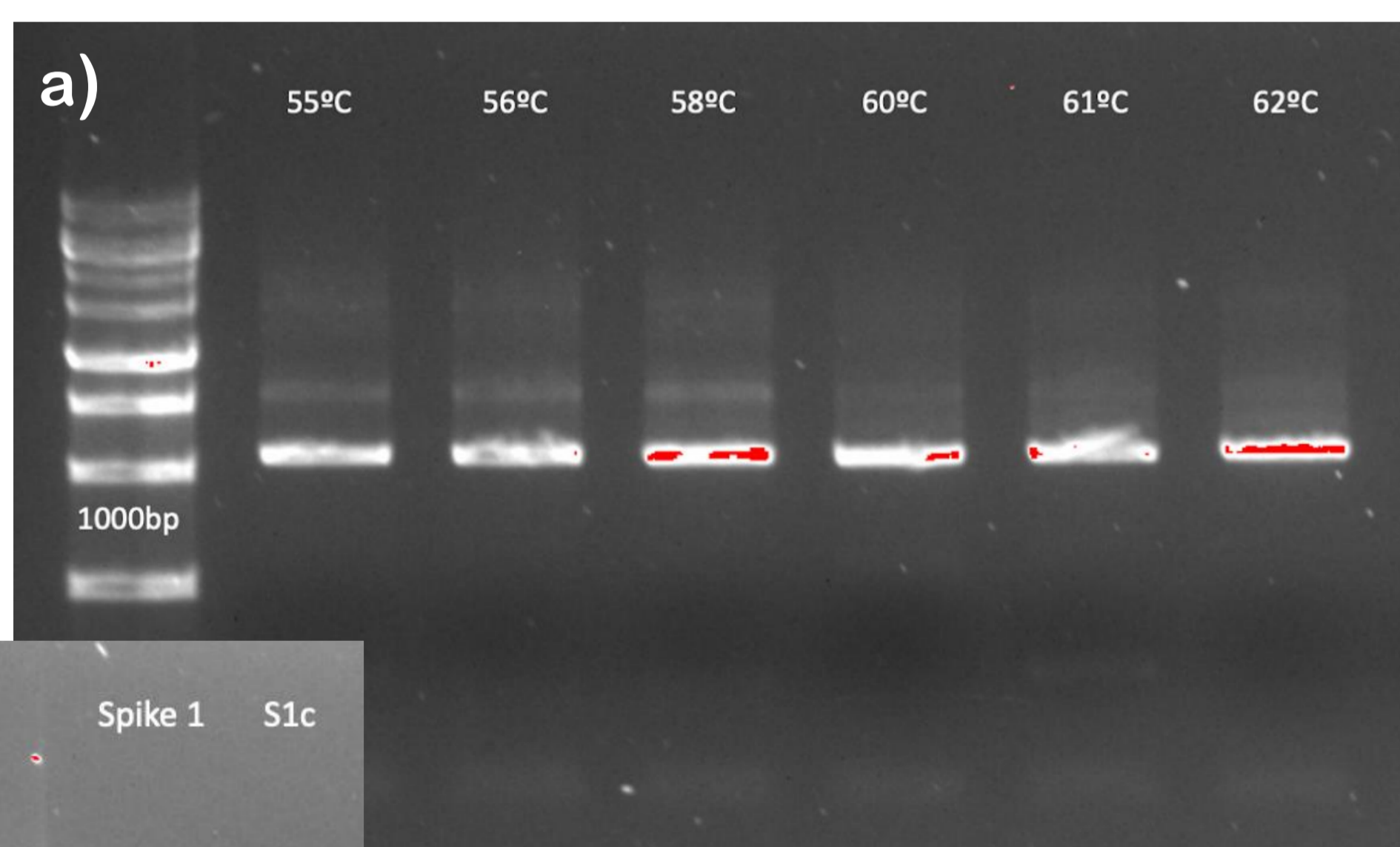


Fig.5- PCR optimization tests using TAQ Phusion polymerase in positive control samples: a) best annealing temperature (62°C); b) DNA detection in diluted samples (S1c=5,83ng/μL)

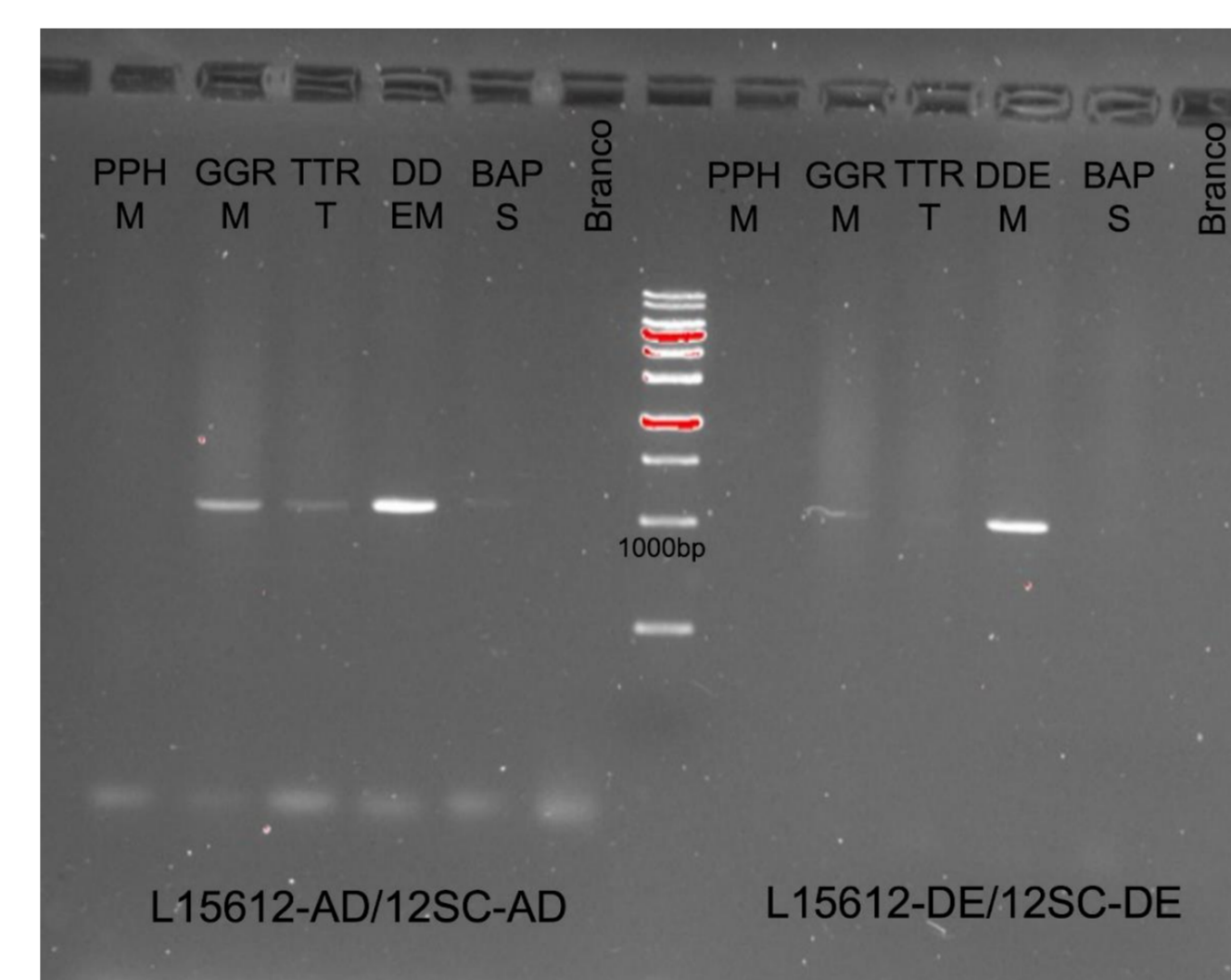


Fig.4- PCR with degenerated (DE) and adapted (AD) primers developed from the original primer set from Zerbini *et al.*, 2007

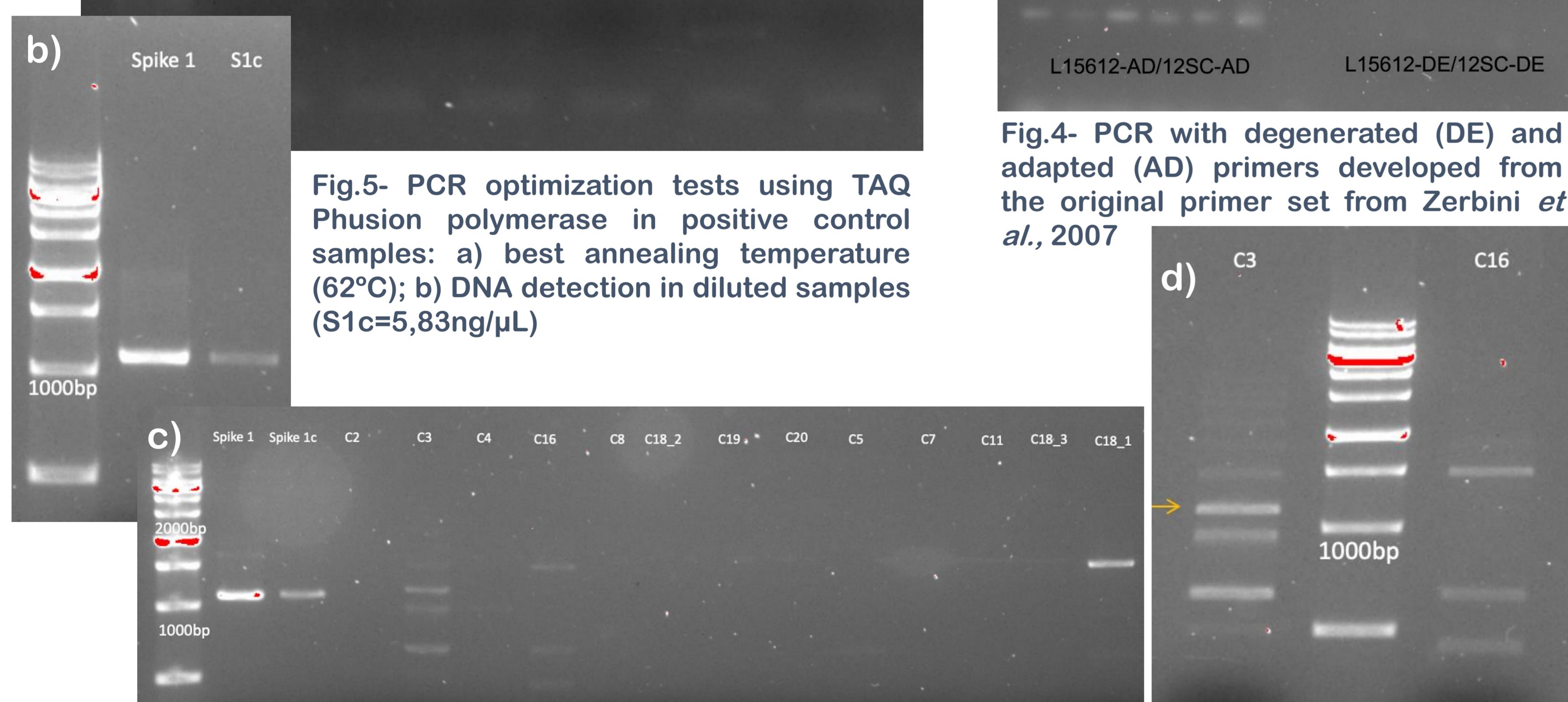


Fig.6- PCR using previously optimized conditions for TAQ Phusion, in eDNA samples collected near the animals at the time of sighting: c) all extracted samples from C2 to C18_1; d) PCR repetition for positive detections

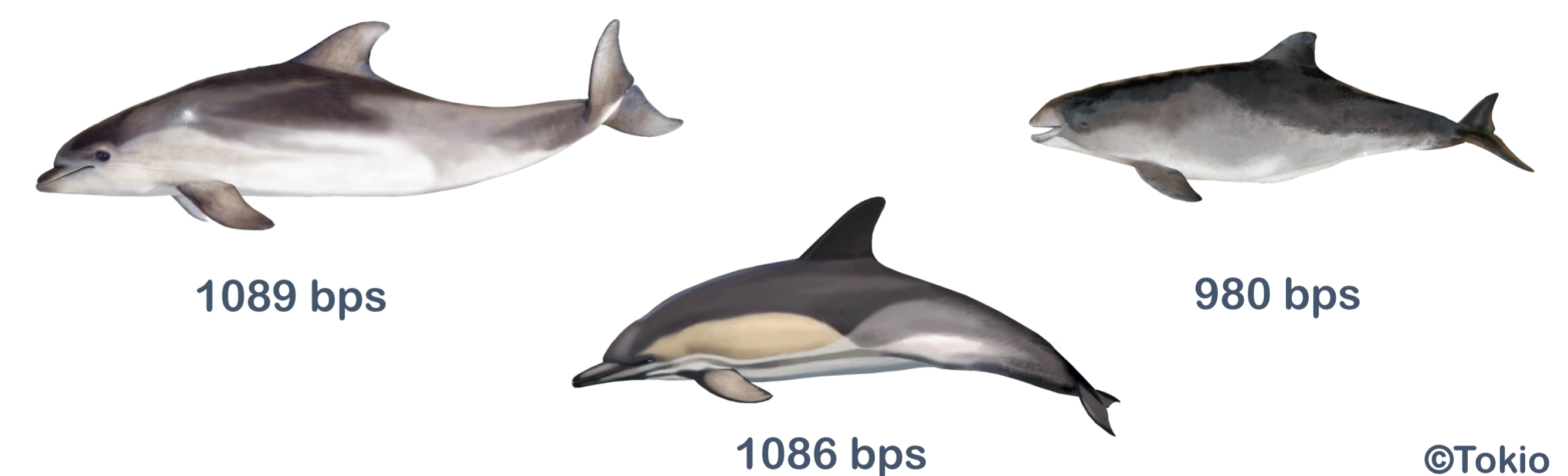


Fig.3- Target amplicon sizes for each of the target species, using the L15812/12SC primer sets, retrieved from Zerbini *et al.*, 2007 [1]

Conclusions

- Factors like salt water do not seem to inhibit the PCR reaction;
- We were not yet able to identify cetacean DNA in environmental samples, probably due to low DNA concentration;
- Although the effectiveness of resorting to eDNA for cetacean monitoring programs remains unclear, these results represent a step forward towards that goal.

Acknowledgements

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References

[1] Zerbini AN, Waite JM, Durban JW, LeDuc R, Dahlheim ME, and Wade PR (2007). Estimating abundance of killer whales in the nearshore waters of the Gulf of Alaska and Aleutian Islands using line-transect sampling. *Mar Bio* 150:1033-1045.

