

Figure S5: Detailed bioinformatic processing of metabarcoding to extract haplotype sequences using the JAMP R package. **A**) Metabarcoding raw data is processed and quality filtered. These steps are integrated in JAMP, but most other standard metabarcoding pipelines could be used as well. **B**) The processed and quality filtered samples from step A would be usually clustered into operational taxonomic units, but are here additionally filtered (retaining reads of only the expected amplicon length and discarding reads of low abundance) and then denoised. **C**) In denoising with usearch unoise3 the strictness of denoising is controlled by the alpha value (low alpha = less noise, bowever more true haplotypes get discarded). **D**) The denoised reads (=haplotypes) are clustered into OTUs grouped by similarity and the abundance of each haplotype for each sample is exported in a table. **E**) The haplotype table is additionally filtered using different thresholds, to reduce the presence of low abundant OTUs and haplotypes and increase data reliability. **F**) The final filtered haplotype table can be used for phylogeographic and population genetic analysis.