# **Appendix**

# Wet-lab methods matter: Extraction and library preparation protocols impact oral microbiome recovery from archaeological dental calculus

A blue and orange grid with black text

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**Figure A.1.** Heatmap showing the species and their relative abundance in the Niger dataset.

A close-up of a chart

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**Figure A*2*.** Heatmap showing the species and their relative abundance in the Hungary dataset. Heatmap was generated with pheatmap (v.1.0.12).

A screenshot of a graph

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**Figure A3.** Fragment length distributions for Niger samples.

A screenshot of a graph

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**Figure A4.** Fragment length distributions for Hungary samples

*.* A group of graphs on a white background

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**Figure A*5*.** Box plots of average fragment lengths for both Niger (A & B) and Hungary (C & D).

A screenshot of a graph

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**Figure A*6*.** Box plots of GC percent for Niger (A & B) and Hungary (C & D) datasets.

A group of graphs showing different results

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**Figure A*7*.** Percentage of unique reads for Niger (A & B) and Hungary (C & D) datasets.

A screenshot of a graph

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**Figure A8.** MapDamage results for Niger dataset when mapped to Anaerolineaceae bacterium oral taxon 439

A screenshot of a graph

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**Figure A9.** MapDamage results for Niger dataset for Methanobrevibacter oralis.

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**Figure A10.** MapDamage results for Niger dataset for Olsenella sp. taxon 807.

A graph of different colored lines

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**Figure A11.** MapDamage results for Hungary dataset for Anaerolineaceae bacterium oral taxon 439.

A screenshot of a graph

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**Figure A12.** MapDamage results for Hungary dataset for Methanobrevibacter oralis**.**

A screenshot of a graph

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**Figure A13.** MapDamage results for Hungary dataset for Olsenella sp. taxon 807.

A red and blue vertical bars with numbers

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**Figure A14.** ChangePoint results. Results indicate that all samples in the Hungarian dataset had a higher frequency of Ts on the 5’ end, as well as a higher frequency of C’s on the 3’ end, than would be expected by chance. The oppose was the case for the Niger dataset. These results are further supported by the SourceTracker2 results.