Genomic Analysis and Environmental Adaptation in the Prehistoric Western Mediterranean

STATE OF THE ART
The **Neolithic** in the western Mediterranean marked major cultural, economic, and social shifts, driven by the advent of farming, animal domestication, and permanent settlements[1]. The region became a dynamic hub, where maritime networks enabled exchanges of goods (obsidian, flint, ivory), ideas, and technologies. During the **Chalcolithic** period, which bridged the Neolithic and Bronze Ages in central and northern Europe, there was a drastic change in the genetic makeup of the population due to the influx of steppe-related peoples[2],[3]. In the Western Mediterranean, however, cultural diffusion dominated with little genetic turnover[4]. However, few human samples have been analysed, and systematic genomics studies of Western Mediterranean populations are lacking. Around 4.2 kya, major climate change brought aridity and cooling[5], leading to desertification, the collapse of agriculture, famine and migration. Climate also affects the spread of infectious diseases by increasing the abundance of vectors[6]. For instance, the Black Death epidemics that occurred during and after the Middle Ages were influenced by climate fluctuations[7].

PURPOSE OF THE PROJECT:
This project investigates the interplay between biological and cultural variation in prehistoric populations, focusing on human–environment interaction from the Neolithic to the Bronze Age (~8000–3000 BP), with emphasis on the Chalcolithic. By analysing individuals from the western Mediterranean, we aim to study genetic diversity, mobility, environmental shifts, and pathogens' roles in the formation of prehistoric societies. Goals include: 1) shed light on the processes that **shaped genetic diversity**, distinguishing between **local movements** (e.g. short-range migrations), **contacts** among the western Mediterranean area, and **socio-cultural transformations**; 2) shed light on the dynamic of **human-environmental interactions** including the impact of pathogens on the historical trajectory of the area 3) understand the demographic impact of catastrophic events, like **climate-driven pandemics**, had on local demography.

MATERIALS AND METHODS
Sample Collection
The project plans to start with ~400 bone/tooth samples from different geographic areas of the Mediterranean basin (~260 from Italy, ~20 from Malta, ~80 from southern Spain, ~40 from southern France). A rigorous quality control protocol will be followed to evaluate the preservation state of the samples[3], with the goal of retaining ~60% of the collection (~240 individuals) for the initial screening.

Wet Lab
All lab work will be conducted in the dedicated aDNA clean lab at the University of Rome Tor Vergata[8]. DNA extraction will target teeth and cochlear regions of petrous bones using modified protocols[9]. Double-stranded genomic libraries[10] will be prepared, indexed, and sequenced on Illumina NextSeq 550 platform at ISIS@MACH. Each library will be sequenced to generate 10–15 million DNA reads. Raw data will be processed via bioinformatic pipelines to assess quality, endogenous DNA content, contamination levels, and genetic sex. Samples with >5% endogenous DNA and good preservation will be selected for a deep sequencing, aiming for genome coverage >0.5X, sufficient for genotype imputation. Based on my experience, we expect ~75% success, yielding ~180 genomes.

Data Analysis
Samples with >0.5X coverage will be imputed[11] and merged with ancient Eurasian genomic data. Analyses will include kinship evaluation, population structure, and identity by descent to explore

genetic relationships and evolutionary dynamics. Reads that do not map to the human genome will be analysed for the presence of potential pathogens by using an already published computational workflow[12]. A positive microbial detection will be defined by specific criteria[13]. Our primary aim is to achieve positive microbial detection; however, if time permits, samples testing positive for pathogens will be further processed using targeted hybridization with myBaits Custom capture kits to obtain full genome coverage[14].

SECONDMENT

I am planning to spend a six-month secondment at the Department of Historical Studies, University of Gothenburg (see attached support letter), from late Year 2 to early Year 3. I will collaborate with Prof. Sabatini to integrate genetic data with archaeological and historical contexts to refine interpretations of prehistoric human–environment interaction and cultural transitions in the western Mediterranean.

Gantt Chart of the project

Tasks	Year 1 I	II	III	IV	Year 2 I	II	III	IV	Year 3 I	II	III	IV
Sampling	▓	▓										
Wet lab	▓	▓	▓	▓	▓	▓	▓					
Screening sequencing		▓	▓	▓	▓	▓						
Deep sequencing			▓	▓	▓	▓	▓	▓				
Data Analysis		▓	▓	▓	▓	▓	▓	▓	▓	▓		
Secondment								▓	▓			
Phd thesis and papers								▓	▓	▓	▓	▓

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