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Monnereau, Aurore, Ughi, Alice, Orecchioni, Paola et al. (11 more authors) (2024) Multiproxy bioarchaeological analysis of skeletal remains shows genetic discontinuity in a Medieval Sicilian community. Royal Society Open Science. 240436. ISSN 2054-5703

https://doi.org/10.1098/rsos.240436

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Cite this article: Monnereau A *et al.* 2024 Multiproxy bioarchaeological analysis of skeletal remains shows genetic discontinuity in a Medieval Sicilian community. *R. Soc. Open Sci.* **11**: 240436. https://doi.org/10.1098/rsos.240436

Received: 17 March 2024 Accepted: 31 May 2024

Subject Category:

Genetics and genomics

Subject Areas:

genetics

Keywords:

ancient DNA, isotope analysis, archaeological science, Sicily, Middle Ages

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Electronic supplementary material is available online at https://doi.org/10.6084/m9.figshare.c. 7313547.

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Multi-proxy bioarchaeological analysis of skeletal remains shows genetic discontinuity in a Medieval Sicilian community

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The medieval period in Sicily was turbulent, involving successive regime changes, from Byzantine (Greek Christian), Aghlabid (Sunni Muslim), Fatimid (Shīʿa Muslim), to Normans and Swabians (Latin Christian). To shed new light on the local implications of regime changes, we conducted a multidisciplinary analysis of 27 individuals buried in adjacent Muslim and Christian cemeteries at the site of Segesta, western Sicily. By combining radiocarbon dating, genome-wide sequencing, stable and radiogenic isotopic data, and archaeological records, we uncover genetic differences between the two communities but find evidence of continuity in other aspects of life. Historical and archaeological evidence shows a Muslim community was present by the 12th century during Norman governance, with the Christian settlement appearing in the 13th century under Swabian governance.

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A Bayesian analysis of radiocarbon dates from the burials finds the abandonment of the Muslim cemetery likely occurred after the establishment of the Christian cemetery, indicating that individuals of both faiths were present in the area in the first half of the 13th century. The biomolecular results suggest the Christians remained genetically distinct from the Muslim community at Segesta while following a substantially similar diet. This study demonstrates that medieval regime changes had major impacts beyond the political core, leading to demographic changes while economic systems persisted and new social relationships emerged.

1. Introduction

Due to its agricultural fertility and strategic position in the central Mediterranean, Sicily has long attracted a diversity of people. During the Middle Ages (5th to 13th centuries CE) Romans, Greeks, Byzantines, Muslims and Latin Christian Northern Europeans competed for control. In 827 CE, the Aghlabid (Sunni Muslim) forces arrived at Mazara from North Africa and by 910 CE had subdued the entire island. From 910 CE, Fatimid Shī'a Muslims from North Africa sought power in Sicily and in 948 CE, the Fatimid dynasty of the Kalbids took control [1]. They created a prosperous province of the Fatimid Empire with its capital at Palermo from 948–1053 CE. In 1061 CE, Norman Christians led by the Hauteville family invaded Messina and established the kingdom of Sicily under Roger II and his successors from 1130–1189 CE [1,2]. In 1194, Swabians led by the Hohenstaufen dynasty took governance of the kingdom, ruling until the death of Frederick II (*stupor mundi*) in 1250 CE.

While these well-documented histories of changing regimes provide a geo-political context for the period, many of the social and economic impacts are far less discernible. The elites who feature in the chronicles were principally seeking political and religious control through military force, wealth and monumentality, but the effects felt amongst the larger population are far from clear. Historical sources indicate that marriage between Muslims and Christians occurred in both Sicily and Spain, although attempts were made to discourage it [3,4]. For example, in 973 CE during the Fatimid dynasty, Muslim traveller Ibn Hawqal observed widespread acculturation in western Sicily, with mixed marriages between Muslim and Christian individuals [5]. The degree to which interfaith marriages occurred during the later Norman/Swabian period remains a question that requires further investigation. Various parameters could be employed to observe this phenomenon, including genetic evidence and the distinctiveness of the cuisines and diets of these individuals, which could serve as useful indicators of social rules.

Human remains provide a direct source of information regarding the lifeways, genetic and cultural affinities of non-elites living through these periods of political turbulence. As such, they provide a powerful, independent source of evidence, enhanced through the application of biomolecular techniques to reveal dietary patterns, residential mobility, genetic diversity and kinship. Using these approaches, here we examine communities of different faiths buried in adjacent cemeteries at Segesta in northwestern Sicily (Trapani Province, figure 1*a*) as it came under Norman and Swabian control during the 12th to 13th centuries CE [6,7]. The Muslim cemetery, situated on the edge of the Greek theatre, consisted of 75 burials, with individuals' faces oriented southeast toward Mecca [6,7]. Approximately 60 m to the southwest, a Christian cemetery was situated on the west side of a church that had been originally constructed circa 1200 CE [8]. The Christian cemetery consisted of multiple burials in 57 stone tombs, with those individuals who were undisturbed lying supine with folded arms [8,9].

By analysing individuals buried under Islamic and Christian rites, we aimed to elucidate the chronological sequence at Segesta and compare aspects of their lifeways, through diet, mobility and genetic ancestry. The correlation between ancestry, diet and faith has rarely been directly examined in historical populations and it is particularly unusual to document the degree of demographic, economic and cultural change during a period of dramatic political transition. To achieve this aim, 27 individuals buried in the Islamic (n = 9) and Christian traditions (n = 18) were selected for multiproxy biomolecular analyses (electronic supplementary material, dataset S1: Genetic and radiocarbon analyses; electronic supplementary material, dataset S3: stable isotopes, electronic supplementary material, dataset S4: compound-specific isotope analysis (CSIA)). All of the 27 individuals were analysed for carbon and nitrogen stable isotope analysis to assess potential dietary differences, 17 for strontium and oxygen to assess levels of mobility, as well as 25 for radiocarbon dating to refine the chronology of the cemeteries. Of the nine individuals from the Muslim cemetery and the 18 individuals from the Christian cemetery, eight and 13 were respectively assessed for genome-wide



Figure 1. Location and chronology of Segesta cemeteries. (a) Site of Segesta indicating the location of the Muslim (C1) and Christian (C2) cemeteries with an inset map showing the island of Sicily with the location of Segesta (1) and the cities mentioned in the text: Palermo (2), Messina (3), and Mazara (4); (b) Chronological model of the radiocarbon dates for Muslim and Christian burials at Segesta using OxCal v4.4 and following the methods described in the Supplemental Information.

screening to investigate genetic similarity, demographic continuity and potential kinship within and between the two cemeteries. A full description of the sampling is provided in electronic supplementary material, information (electronic supplementary material, dataset S1, dataset S3).

2. Material and methods

2.1. Radiocarbon dating

Detailed information about the burials and individuals can be found in the electronic supplementary materials (electronic supplementary material (SI) text §1, dataset S1 and dataset S3). Twenty-five of the samples were radiocarbon-dated using accelerator mass spectrometry (AMS) at SUERC, the University of Glasgow. Radiocarbon results were calibrated with the IntCal20 atmospheric curve [10] using OxCal online version 4.4; reservoir effects were applied where appropriate when undertaking Bayesian chronological modelling to examine whether the cemeteries were used at the same time [11,12] (see electronic supplementary material text §2, dataset S1, dataset S5).

2.2. aDNA analysis

Ancient DNA analysis was undertaken to examine the genetic diversity and kinship of the Segesta population. Twenty-one individuals from Segesta were sampled for aDNA analysis. The petrous portion of the temporal bone was preferentially selected in order to maximize the recovery of human endogenous DNA. If a petrous was not available, then either a tooth or a long bone was selected [13]. The ancient DNA laboratory experiments were conducted within the ancient genomics laboratory at BioArCh, University of York, UK; detailed methods are presented in the electronic supplementary material, text §3. DNA was extracted from skeletal elements, converted into double-stranded Illumina libraries [14], and sequenced on an Illumina HiSeq platform at the GeoGenetics Sequencing Core in Copenhagen, Denmark and Novogene in Sacramento, California, USA (electronic supplementary material, text §3). Mitochondrial DNA was enriched in two samples with low endogenous DNA content using a myBaits hybridization capture kit (Arbor Biosciences, Ann Arbor, Michigan, USA) [15]. Samples which exhibited the expected patterns of DNA degradation [16] and demonstrated no contamination in the mitochondrial genome [15] and X chromosome [17] were included in further analyses. Using a minimum mapping quality of 30 and base quality of 30, uniparental haplogroups were determined by using HaploGrep [18] and Yleaf [19]. Ancient DNA data from Segesta and Himera [20] were merged with published data for the Human Origins (HO) dataset and archaeological individuals compiled in the Allen Ancient DNA Resource (V50) [21]. To avoid the impact of uncorrected DNA damage, analyses excluded transition sites. A pseudo-haploid approach was implemented for nuclear SNP loci, using the random sampling of one read, and then using EIGENSOFT [22,23] for PCA analysis and ADMIXTURE [24] to infer ancestry components. To estimate affinities among populations, we calculated outgroup- F_3 -statistics using ADMIXTOOLS software option 'qp3pop' and 'inbreed:yes' [25].

2.3. Isotope analysis

Full details of stable and radiogenic isotope analysis are presented in the electronic supplementary material, information (electronic supplementary material, text §4 and §5). Bone collagen extraction was carried out according to established protocols, including an additional ultrafiltration step [26,27]. Collagen from adult humans (n = 11) and terrestrial and marine fauna (n = 21) was selected for compound-specific stable isotope analysis based on the samples with the best preservation. Collagen was prepared for GC-C-IRMS, following hydrolysis to release amino acids. For these, δ^{15} N measurements were carried out on at least nine individual amino acids using the approach previously described by Soncin *et al.* [28]. Quality control criteria are described in electronic supplementary material text §4 and §5. The δ^{18} O values of tooth enamel carbonates were determined following the procedure described by Miller *et al.* [29], while tooth enamel was prepared for 87 Sr/ 86 Sr measurements following the procedure described by Leggett *et al.* [30].

3. Results and discussion

3.1. Chronology, kinship and ancestry

We examined the chronological relationship between the burials at the two cemeteries to establish whether they were drawn from communities living contemporaneously at Segesta. Direct AMS dating of bone collagen from 25 individuals, 9 from the Muslim cemetery and 16 from the Christian cemetery, indicates that the former began before the latter (figure 1b; electronic supplementary material, dataset S1). Bayesian chronological modelling estimates the Islamic burials began between 770–990 CE (95% probability; figure 1b) and ended by 1190–1380 CE, while the Christian burials started between 1140-1220 CE and concluded by 1270-1330 CE. A formal comparison of the posterior distributions indicates there is a 99.3% probability the dated Christian burial activity began before the end of the Muslim burial activity, from which we conclude that both communities likely resided concurrently for some period of time in the early 13th century. These results complement archaeological evidence that after the arrival of the Christian newcomers, Muslims continued to live nearby. For example, structures found away from the Norman castle at Segesta employed ground plans and building techniques of rough-cut stones bonded with earth, forms used in the superseded Muslim phase at the castle site [31]. In a similar manner, excavations of occupation levels in Area SAS 5 led to the discovery of coinage of both Henry VI (1194–1197) and rebel leader Muhammad Ibn Abbād (c. 1220) [31,32].

We evaluated genetic sex and kinship in the individuals using DNA analysis of the skeletal material. Endogenous human DNA was successfully extracted from 21 of the AMS-dated individuals, accounting for 0.3–42.6% of the obtained sequences (mean = 15.4%) and yielding a depth of coverage on the nuclear genome of 0.00–1.85× (mean = 0.48×; see electronic supplementary material, dataset S1). DNA damage profiles and contamination estimates were consistent with degraded DNA with minimal human contamination (electronic supplementary material, figure S1; dataset S1). Genetic sex determinations [33] were largely in agreement with the osteological assessment of the adult skeletons, except for SGBN9 (electronic supplementary material, dataset S1), who is presumed to have been misidentified osteologically. Genetic sexing of non-adults showed that one child from the Muslim cemetery was genetically male (XY), one child from the Christian cemetery genetically female (XX), and the other seven children buried in the Christian rite were genetically male (XY). To identify biological kinship relationships within and between the cemeteries, we explored uniparental markers (mitochondrial DNA and Y-chromosomal haplotypes) as well first, second, and third-degree relationships through nuclear single nucleotide polymorphisms (SNPs) using READ software [34]. Although a number of shared mitochondrial DNA and Y-chromosomal haplogroups highlighted potential biological relationships within both cemeteries, familial relationships were only confirmed using nuclear DNA within the Christian cemetery. These were identified first-degree relationships (i.e. parents/offspring or siblings) between individuals SGBN18/SGBN19 and SGBN20 (electronic supplementary material, dataset S1)—all male infants interred in Tomb 17, and with similar dates (SGBN18: 1215–1365 cal CE; SGBN19: 1195–1305 cal CE; SGBN20: 1225–1385 cal CE). As the sequences were obtained from three complete petrous bones, the combined genetic and osteological data suggest that SGBN18 and SGBN19 (right and left petrous) were likely to have been the same individual or twins (hereafter jointly analysed as SGBN18_19), whereas SGBN20 was a sibling. No further relationships (i.e. second-or third-degree) were discernible.

We next explored uniparental genetic markers. Most individuals carried mitochondrial DNA haplotypes which are widely distributed across Eurasia; however, SGBN2, a male buried in the Muslim cemetery carried mitochondrial DNA haplotype L3e5 which is primarily found in sub-Saharan Africa (see details in electronic supplementary material, text §3 and dataset S1). The Y-chromosome haplotypes were suggestive of differences between the cemeteries: individuals from the Muslim cemetery carried haplotypes associated with North Africa (E1b-M81 and E1b-M310.1) [35,36] and the Eastern Mediterranean (J2b-M241) [37], while four of the nine individuals buried in the Christian cemetery belonged to haplogroup R1b-M269, a haplogroup which is primarily found today in Western Europe [38].

To undertake a more detailed investigation of ancestry, we analysed the genome-wide data where at least 10 000 transversion SNPs overlapped with the Human Origins SNP panel used in human palaeogenomics [39]. Principal components analysis (PCA) was performed by projecting archaeological individuals onto the diversity of modern populations available in the Allen Ancient DNA Resource (AADR) [21] (electronic supplementary material, dataset S1, dataset S2), using three geographic levels of inquiry. Against a worldwide panel of 141 modern populations, we observed that all Segesta individuals fell within the PCA-space represented by Europe and North Africa, except for SGBN2, who fell within the diversity of sub-Saharan African populations (electronic supplementary material, figure S2). Constricting the analysis to modern Eurasian and North African populations, we observed individuals from the Muslim cemetery showed affinity to one another, plotting between modern populations from Southern Europe, Southeastern Europe, North Africa and the Near East, the latter set including Jewish populations from the Near East and North African (figure 2a and electronic supplementary material, figure S3). One individual, SGBN7, plots between Near Eastern populations and modern North African populations. The individuals buried in the Christian cemetery plot separately in PCA-space, generally situated near modern populations from Eastern, Southern, Southeastern and Western Europe. Those results indicate a genetic distinction between the groups, with no examples of individuals in one cemetery having a stronger genetic affinity to those from the other cemetery. While the available assemblage was biased toward males, the number of children from the Christian cemetery provided an opportunity to identify offspring of interfaith unions: none were detected. Continuing at the scale of Eurasia and North Africa, we also evaluated these groups' affinity to Iron Age Sicani individuals from Sicily [20], finding the Christian individuals overlapped the PCAspace occupied by the ancient Sicilian Iron Age individuals (electronic supplementary material, figure S3). When SGBN2 is examined in the context of African populations, we observe the individual plots with groups from West and East Africa (figure 2b; electronic supplementary material, figure S4).

The contrast in ancestry between individuals buried in the Islamic tradition compared to those afforded a Christian burial rite was further examined with ADMIXTURE and *F*-statistics. ADMIXTURE analysis indicated that ancient Moroccan and west sub-Saharan ancestries are mainly present within Segesta individuals buried in the Islamic rite (figure 2*c*, electronic supplementary material, dataset S1). We applied an outgroup- F_3 -statistic (X, modern/ancient populations: Jul'hoan_North) to explore population affinities between the ancient Segesta individuals buried under Christian rite, Islamic rite and previously published ancient and modern populations (electronic supplementary material, dataset S2), however, no significant affinities were determined, likely as a result of low numbers of transversions in the dataset (electronic supplementary material, text §3; figure S5–S8).

3.2. Lifeways, diet and mobility

Next we set out to determine whether there were any discernible differences in dietary lifeways at Segesta and, if so, whether these correlate with faith and ancestry. The investigation of diet through stable isotope



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Figure 2. Genetic diversity at Segesta. (a) PCA showing Segesta samples (greater than 10 000 transversion SNPs) against a panel of modern Eurasian and North African populations. PC1 is presented on the y-axis as it follows a north-south gradient; (b) PCA showing SGBN2 individual against a panel of modern African populations; (c) unsupervised ADMIXTURE plot, assuming K = 8 ancestral populations.

analysis reflects long-term habitual attitudes to food as well as economic practice and social status, all of which may vary culturally in a single locality. The approach has been employed to demonstrate differences in diet between individuals of different faith groups (e.g. Christians and Muslims) in medieval societies [40,41]. For example, differences in the bone collagen stable nitrogen (δ^{15} N) and carbon (δ^{13} C) isotope values were noted within the multi-faith community at Gandia on the Mediterranean Iberian coast, dating to the 13th–16th centuries CE, with Muslims consuming a greater amount of marine fish and C_4 plants (e.g. millet, sorghum and sugarcane) in their diet compared to Christians, perhaps reflecting socially restricted access to resources, such as terrestrial animals and C₃ plants (cereals, legumes and pulses at this time [41].

At Segesta, collagen was extracted for stable isotope analysis from a slightly larger set of human remains compared to those selected for AMS dating and genomic analysis, partly in order to more reliably compare the populations but also due to sample availability. Notably, infant diets may vary more widely and partially reflect the effects of breast-feeding [42]. The petrous bone, preferentially targeted here for enhanced aDNA preservation, may also reflect infant dietary practices due to its relatively lower turn-over rates [43]. Significantly, there were no significant differences between bulk δ^{15} N and δ^{13} C of collagen from adults buried under Islamic and Christian traditions (figure 3a,b, electronic supplementary material, dataset S3), such as one may expect due to habitual differences in diet. All the individuals bar one (SGBN24A) had diets dominated by C_3 plants or C_3 foddered terrestrial animals, with no significant enrichment in collagen ¹³C (i.e. >1‰) compared to comparative values for 10th-13th centuries terrestrial fauna from Mazara [44]. While a lack of C4 cereals (millet and sorghum) is expected during this phase in Sicily, as reflected in archaeobotanical assemblages from across the island [47,48], the apparent absence of marine foods is interesting given the site's proximity to the coast (ca. 10 km).

To investigate diet further, we deployed a higher resolution compound-specific approach and measured the δ^{15} N values of individual amino acids hydrolysed from the collagen of 13 adult individuals: five from Islamic and eight from Christian burials. Amino acids can be traced to dietary sources with greater certainty than bulk bone protein, allowing the contribution of dietary sources to



Figure 3. Dietary isotope analysis. (*a*) Scatter plot of bulk stable carbon (δ^{13} C) and nitrogen (δ^{15} N) isotope data from humans from Segesta separated by burial rite (Ch = Christian, Is = Islamic), age (A = Adult, N-A = non-adult) including published animals for comparison (Herbivores, Omnivores and Marine fish, mean $\pm 1\sigma$) from Mazara [44]; (*b*) Bagplot comparison of δ^{13} C and δ^{15} N values for adults from Segesta separated by burial rite, with data distributions indicated on the margins; (*c*) Collagen compound specific stable isotope data plotted as the δ^{15} N of glutamic acid against the δ^{15} N of phenylalanine. The trophic position lines (T = terrestrial trophic position, M = marine trophic position) shown are from Naito *et al.* [45] for reference only [see 46].

be quantified at greater precision and accuracy [e.g. 28]. Of these, the δ^{15} N values of phenylalanine (Phe) and glutamic acid (Glu) have been used to better determine trophic position and the degree of aquatic protein consumption [49]. Even using this higher resolution approach, there were no discernible differences between individuals buried according to the different faiths (figure 3*c*); the majority of individuals have Glu/Phe δ^{15} N spacings (Δ^{15} N_{Glu-Phe}) indicative of diets dominated by terrestrial plant/animals, with just a single individual (SGBN15, a Christian burial) having greater access to marine foods during their life. Using this approach we were are also able to confirm that the ¹³C enrichment noted in the collagen of SGBN24 most likely derived from the consumption of C₄ plants, or C4 fed animals, rather than marine foods due to its relatively low estimated trophic position (Δ^{15} N_{Glu-Phe} ~ 5.7‰).

We also obtained information regarding residential mobility by undertaking strontium (Sr) and oxygen (O) isotope analysis of selected individuals where dental samples were available [50] (figure 4). By comparison with the predicted local ranges of both isotopes for Sicily [51–53] there appears to be no convincing evidence of extensive mobility in either community. Individuals from the Christian cemetery have a wider range of Sr isotope ratios, which may indicate, collectively, that they originated from a broader geographic area. One individual buried in the Christian cemetery (SGBN24A) was an Sr outlier and also had a greater proportion of C₄ foods in their diet, offering convincing evidence of a non-local. With this exception, the remaining Sr ratios reflect values that are within the range of Sicily [51] but also shared in many regions across the Mediterranean and indeed Europe, so some level of migration cannot be ruled out in either period. All δ^{18} O isotope values are also in keeping with other Southern Mediterranean values and indeed other measurements of other ancient individuals from Sicily [53]. Notably, however, individuals from the Christian cemetery possess higher δ^{18} O values compared to individuals from the Muslim cemetery, which points to some differentiation in their origin or, perhaps more likely given the lack of difference in their Sr values, access to different sources of drinking water.

3.3. Early medieval Segesta: a multi-faith community

The combined historical, archaeological and biomolecular research applied to the populations buried at medieval Segesta has allowed us to elucidate community and interfaith dynamics. Based on the results of our multi-proxy analysis, we propose that if the communities at Segesta were using the respective cemeteries concurrently during the late 12th and early 13th centuries—as seems highly probable from the archaeological finds and dates available—there was little to no interfaith marriage or biological relationships between the individuals we analysed who were buried in Islamic and Christian rites. Christian burials are also apparent throughout the period of Muslim control of Sicily. At Segesta, the genetic data from the period of Swabian control of the island are consistent with a lack of genetic homogeneity between the communities, with closer affinities to modern Europeans for the Christians.



Figure 4. Mobility isotope analysis. Plot of strontium (left) and oxygen (right) data for tooth enamel from humans from Segesta separated by burial rites (Ch = Christian, Is = Islamic). Dashed lines represent predicted local values, for Sr [51], modern drinking water converted to VPDB [52] and published oxygen data from 5th century BCE individuals from Himera [53].

Interestingly, the Sr and O data suggests that with one exception, there is little evidence the Christian individuals resided outside Sicily during their childhood, thus they may have descended from immigrants of previous generations. Alternatively, this local signal could result from continuity of a Christian population in Sicily from the Iron Age to the Norman period, however, a larger sample size of individuals from multiple archaeological sites would be needed to explore this hypothesis.

The genetic data from Segesta might appear surprising given the length of time that Muslim and Christian communities co-existed on Sicily. Although Norman governance was in force in Sicily from the mid-11th century and Swabian governance from the end of the 12th century, this is not reflected in the archaeology of the burial rites. Radiocarbon dating at Segesta has shown that burial in the Islamic rite was practised from the 9th century to the 13th, only ceasing after the mid-13th century when many Muslims were deported under Frederick II [1,54,55]. Even if we take into consideration difficulties in distinguishing Arab, Berber populations, who may share considerable ancestry, the majority of individuals afforded Christian burials at Segesta date to over a century following the Norman invasion yet remain clearly distinguishable from those in Islamic burials.

Moreover, the proximity of the burial grounds and settlement areas within the former archaic city implies that Muslims and Christians occupied adjacent spaces and had similar access to markets. This view is supported by the study of personal names and land records in Norman times showing that Muslim and Christian communities coexisted in town and country through the Islamic and Norman period [56]. They resided in different quarters of the town and buried in separate cemeteries, but had social relations [1,5,55]. For instance, in 1184 CE, Spanish Muslim traveller Ibn Jubayr observed instances of intermarriage between Muslims and Christians, primarily unions between Muslim men and Christian women. He also reported that Muslims in the countryside lived together with Christians on their estates and were treated well by them [1,5,55]. Christian women in Palermo

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dressing like Muslims and speaking fluent Arabic [1,5]. It seems that a social divide between the faiths became 'unequivocal' only between Muslims and the later Latin Christians [1] appears to be what is reflected by the genetic results at Segesta.

In contrast to lack of evidence for intermarriage, the carbon and nitrogen isotope data shows that both groups had access to foods with similar isotopic values, with no privileged access to commodities, such as fish or meat, which are easily distinguishable using this approach and have been observed elsewhere [41]. Complementary evidence suggests that these were not just broad dietary similarities, but rather that there were shared culinary practices during this period. In particular, the 12th-century ceramic assemblage at Segesta was noted by the excavator to imply a certain hybridity in their use, in which local and Islamic types such as filter jars and hand-made cooking pots occurred with more exotic individual plates and bowls [31]. At a wider scale, emerging evidence from chemical analysis of pottery residues from across Sicily shows continuity in the types of foods prepared across periods of Islamic to Norman and Swabian control [57]. While diet and culinary practices represent two proxies of more complex lifeways, they are nevertheless strongly linked with occupation, location of residence and religious affiliation [58]. In Sicily, although Christian political control discouraged interfaith marriage, our data and historical observations of individuals like Ibn Hawqal and Ibn Gubayr suggest that cultural aspects of life were shared between communities.

4. Conclusion

This analysis at the site of Segesta provides a unique glimpse into the interactions between religious communities during the Middle Ages, made possible by the long period in which Muslims and Christians co-existed in Sicily (9–13th centuries CE) and the chance of investigating a site at which Muslim and Christian cemeteries lay adjacent and overlapped chronologically. Funerary archaeology provides powerful statements of tradition and cultural and religious affiliation, but less often reveals the details of everyday life. New insights were achieved here using biomolecular investigations. Isotopic analyses indicated individuals enjoyed a similar diet with little evidence for extensive mobility, however, the genetic analyses indicate that the Muslim and Christian individuals were not only separated by the location of their burials but also by their genetic heritage with no evidence of kinship between the two communities. Based on the assemblage analysed here, our results suggest that the two communities at Segesta could have followed endogamy rules, however, this should not be taken as generally applicable to Medieval Sicily without further analysis of other contemporaneous sites. In this sense Segesta provides a snapshot of the later Swabian period arrivals that has yet to be repeated elsewhere.

Ethics. This study follows ethical regulations from the country of provenance (Italy) as well as ethical considerations throughout sampling and destructive analyses. The Authorities responsible for archaeological resources, granted the permissions needed to analyse the ancient human individuals from Segesta for bioarchaeological analysis. **Data accessibility.** The genetic data files are available in NCBI Project PRJNA1082095. All other data is available within the electronic supplementary information [59].

Declaration of Al use. We have not used AI-assisted technologies in creating this article.

Authors' contributions. A.M.: conceptualization, data curation, formal analysis, investigation, visualization, writing original draft, writing—review and editing; A.U.: conceptualization, data curation, formal analysis, investigation, visualization, writing—review and editing; P.O.: data curation, resources, writing—review and editing; R.H.: investigation, writing—review and editing; H.M.T.: formal analysis, investigation, writing—review and editing; E.N.: formal analysis, funding acquisition, investigation, supervision, writing—review and editing; D.H.: formal analysis, investigation, visualization, writing—review and editing; P.LR.: formal analysis, investigation, writing review and editing; A.Mol.: conceptualization, funding acquisition, project administration, writing—review and editing; M.C.: conceptualization, funding acquisition, project administration, supervision, writing—original draft, writing—review and editing; C.F.S.: conceptualization, supervision, visualization, writing original draft, writing—review and editing; C.F.S.: conceptualization, supervision, writing—original draft, writing—review and editing; N.M.A.: conceptualization, formal analysis, supervision, visualization, writing—original draft, writing—review and editing; N.W.: conceptualization, formal analysis, investigation, supervision, visualization, writing—original draft, writing—review and editing; N.W.: conceptualization, formal analysis, investigation, supervision, visualization, writing—original draft, writing—review and editing; N.W.: conceptualization, formal analysis, investigation, supervision, visualization, writing—original draft, writing—review and editing; N.W.: conceptualization, formal analysis, investigation, supervision, visualization, writing—original draft, writing—review and editing.

All authors gave final approval for publication and agreed to be held accountable for the work performed therein. **Conflict of interest declaration.** We declare we have no competing interests.

Funding. This project has received funding from the European Research Council (ERC) under Horizon 2020 research and innovation programme Grant agreement No. 693600, 'The Archaeology Of Regime Change: Sicily In Transition' (SICTRANSIT). The strontium isotope analysis was supported by the European Regional Development **Acknowledgments.** We thank the Danish National High-throughput Sequencing Centre for assistance in generating sequencing data. This project was undertaken on the Viking Cluster, which is a high performance compute facility provided by the University of York. We are grateful for computational support from the University of York High Performance Computing service, Viking and the Research Computing team. We would also like to thank the Segesta Archaeological Park and Dr. R. Di Salvo for kindly allowing us to take samples from the skeletons of the two cemeteries. We also thank the three anonymous reviewers for their insightful comments and helpful suggestions.

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