

RESEARCH ARTICLE

Population dynamics in pre-Inca human groups from the Osmore Valley, the Azapa Valley and the coast of the South Central Andes

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Abstract

The present study applies a dental morphological perspective to the understanding of the complex pre-contact population history of the South Central Andes, through the detection of the underlying dynamics, and the assessment of the biological ties among groups. It presents an analysis of 1591 individuals from 66 sites that date from the Archaic to the Late Intermediate phases from Bolivia, Chile and Peru. The results suggest this area is characterized by significant movement of people and cultures and, at the same time, by long standing population continuity, and highlight the need for wider perspectives capable of taking into account both the different micro-regional realities and the region in its entirety.

Introduction

Complex population dynamics have characterized settlement patterns and migrations in the South Central Andes making this vast area, encompassing western Bolivia, southern Peru and northern Chile, of particular bioarchaeological significance. Since 12,000 BC [1] the area was inhabited by populations practicing fishing economies. Between 8,000 and 1,000 BC, these slowly gave way to settlements that relied on plant and animal husbandry [2]. This led to the development of more centralized forms of power and to the rise of important polities—such as the Wari and the Tiwanaku—that extended their authority through a variety of different strategies: colonization by agropastoral migration in the case of Tiwanaku and centralized expansion

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in that of Wari [3–5]. Though consensus has yet to be reached regarding these migrations and their influence on the populations that inhabited the region, there has been intense academic debate that has also resulted in the formulation of different hypotheses regarding Tiwanaku, its extent, and the impact of its colonies in various valleys in the region. One may state that Tiwanaku was likely founded in the southern Titicaca Basin, immediately south of Lake Titicaca, during the first or second century AD, and that it thrived, becoming the capital of an archaic state from around AD 500–1000 [6]. Tiwanaku reached its apogee during the Middle Horizon period, around AD 700 [7], and began its decline by AD 900 [8]. Archaeological evidence suggests it expanded its sphere of influence by establishing permanent colonies in many surrounding territories [3, 9] among which the Osmore–or Moquegua–Valley in southern Peru [10] and the Azapa Valley in northern Chile [11–16].

The Wari, on the other hand, appear to have implemented a mosaic pattern of control over their vast territories [4] by implementing both a militaristic expansion [5] as well as a more diplomatic approach [4]. They integrated the valleys and the regions surrounding their core into a broader system of political control using them as gateways to the coast and exploiting them for agricultural production to satisfy the needs of their vast and expanding population.

The growing number of human skeletal collections available has led to extensive analyses of the cultural, genetic and ethnic changes that took place in these valleys in pre-Inca times [7, 17–26]. According to Varela and Cocilovo [15], the population in the Azapa Valley, originally comprised of Archaic fishermen, changed after the arrival of settlers likely migrating from the circum-Titicaca area during the Early Intermediate (Formative) period. Though Varela and Cocilovo [15] state the migrations appeared to have had greater impact on the populations that lived in the valleys compared to those that lived on the coast, they believe there was substantial gene flow between these two regions throughout the period. They hypothesize that, in part because of the strong influence exerted on the valley by the Tiwanaku, as of AD 300, these two populations started to change and diversify. The model depicted in their study indicates an increase of regional gene flow to the valley throughout the Tiwanaku period (AD 300–800) that, in combination with the gradual reproductive isolation of the coastal populations, led to the progressive differentiation of these previously very similar groups. It is only during the Late Intermediate–after the decline of the Tiwanaku State between AD 800–1,000 and possibly in consequence to it–that Varela and Cocilovo [15] detect a substantial decrease of gene flow to the valley. In contrast, Sutter and Mertz [26] believe there is no direct evidence indicating the Tiwanaku replaced the Azapa groups during the Middle Horizon. Their study indicates continuity through time in the valley and suggests that the similarities with the Tiwanaku are, most likely, the effect of interaction and cultural exchange rather than the product of intense immigration into the valley from the northeastern regions south of the Titicaca Basin. In a later study, Sutter [24] detected biological continuity among coastal groups as well as among inland groups in the Azapa Valley that was not accompanied by significant, if any, continuity between these two regions; thus, further supporting his theories that refute significant influence of allochthonous populations to the local gene pool.

The most widely accepted hypotheses regarding the population history of the Osmore Valley indicate that ethnically diverse colonists from the Tiwanaku State spread and settled into different areas of the Upper Valley, where they established permanent outposts such as Chen Chen and Omo [3, 27, 28]. Settlement pattern data from the Osmore Valley [3] suggest that the Tiwanaku expansion proceeded at a massive demographic scale and through direct colonization. According to this hypothesis, the colonists settled into four different enclaves, avoided transculturation with the valley's indigenous inhabitants [19, 27] and continued to receive a small influx of individuals from different parts of the Tiwanaku polity through time [29]. Regardless of the extremely strong influence it had on the valley, Tiwanaku does not appear to

have ever exerted complete territorial control. On the other hand, Tiwanaku seemingly coexisted, presumably peacefully [28], with both the indigenous inhabitants and, as of AD 600, with the Wari outposts in the region such as Cerro Baúl [30–32]. These studies indicate that in spite of the close proximity (at times of no more than 10–20 km) and the long period of time throughout which they must have necessarily interacted, intercultural transmission in the valley was extremely limited and each of these communities maintained its own traditional customs, architectures and technologies [27].

Even though widely accepted, these hypotheses regarding the population history of the Osmore Valley are by no means the only ones that have been advanced. In fact, Mosely et al. [33] believe the Tiwanaku population in the Osmore Valley could have been an autonomous group that, under the Tiwanaku sphere of influence [17], adopted by means of cultural transmission the Tiwanaku customs and way of life for, as stated by Blom, “. . . the mere possession of these (Tiwanaku) artifacts might not indicate population movement and shifts in identity.” [17: p.155].

Given that the presence of shared material culture may indirectly suggest biological contact between different entities or groups, but it may not demonstrate it or provide direct evidence of population admixture, population studies have, in the past years, shifted towards multidisciplinary approaches that include bio-archaeological analyses based on evolutionary theory. These have provided great insight into the population dynamics of the region but have, to this day, mainly focused on specific research questions or areas [17–19, 26, 28, 34–38]. Exceptions to this are a number of studies conducted by Sutter [2, 7, 24] that broaden the perspective by taking into account an area that includes both the Osmore and the Azapa Valleys. These studies analyze, through different means, the complex settlement histories that characterize each of the valleys in order to better understand the region as a whole as well as the ties between the different populations that inhabited it. The results indicate that the situations depicted in the different areas are far from homogeneous and that, while there are indications that the Late Intermediate inhabitants of the Osmore Valley may derive from immigrant populations, the inhabitants of the Azapa Valley appear to derive from in situ microevolutionary processes. These differences certainly indicate that much is yet to be understood regarding the population history and the biological ties between the different groups that inhabited both the valleys and the *altiplano* [39], and they reaffirm the necessity of large-scale multidisciplinary approaches.

Given the centrality of the Tiwanaku to the population history of the region we believe the area under investigation needs to be broadened even further to include the southern portion of the Titicaca basin and incorporate true Tiwanaku samples as well as their supposed outposts to the valleys. We feel this is especially crucial given the results obtained by us in a previous dental morphological study [20] that suggest caution in considering the inhabitants of the outposts to the valleys representative of the presumed ancestral population, especially when dealing with complex peopling scenarios as is the case in the South Central Andes. The study was a smaller scale analysis of the pre-Inca inhabitants of the Osmore Valley that compared Formative, Middle Horizon and Late Intermediate samples (including the supposed Middle Horizon Tiwanaku outpost of Chen Chen). The analysis detected affinities between the Chen Chen and the Wari affiliated samples suggesting that either the inhabitants of the Tiwanaku colony of Chen Chen had more ties than expected to the earlier autochthonous Wari groups of the region or that perhaps the very attribution of Chen Chen as a Tiwanaku colony should be questioned.

In the present study we wish to understand the ties among the culturally diverse human groups that inhabited the region from Archaic to Late Intermediate times with particular focus on the Tiwanaku, the manner in which they colonized new territories and the ties they established in both time and space with the neighboring communities. We do so by including in the

study data pertaining to a small but central sample formed of 35 Middle Horizon individuals from Tiwanaku as well as from 199 Late Intermediate individuals from the circum-Titicaca area.

Materials and methods

The present analysis comprises dental morphological data pertaining to 66 Peruvian, Bolivian and Chilean pre-Inca sites— 1591 individuals—pooled in 15 groups, on the basis of the chronology, location and cultural affiliations attributed by the museums where the collections were curated (Fig 1, Table 1 and S1 Table). The data pertaining to the Middle Horizon Wari and Tiwanaku affiliated sites and the Late Intermediate sites of Chancay and Titicaca (790 individuals pooled in 5 groups) were scored directly by one of the authors of the present study (Argañini, Candilio or Coppa). The dataset thus gathered was then combined to that from a published dental morphology study pertaining to the Peruvian Late Intermediate sites of Chiribaya, Yaral and San Geronimo and Chilean sites from the Azapa Valley [23].

Permits were obtained for all aspects of the study and were issued by the following authorities (listed alphabetically): The American Museum of Natural History, New York, USA; the Anthropology Museum “S. Sergi”, Sapienza University, Rome, Italy; the Leverhulme Centre for Human Evolutionary Studies, Duchworth Foundation, Cambridge University, UK; the Musée de l’Homme, Muséum National d’Histoire Naturelle, Paris, France; and the Museo Antropológico de la Escuela Profesional de Antropología de la Universidad Nacional del Altiplano, Puno, Peru. The inventory provided in S1 Table indicates what sites were sampled in each of these institutions while the supplementary tables (S7, S8, S15 and S16 Tables) indicate what samples, or repository numbers, were analyzed in each of these institutions.

Raw data for the 79 morphological traits scored is provided in S2 to S16 Tables whereas the pooled data attained by combining the data scored to that obtained in literature [23] is listed in S17 Table.

Data were collected using the Arizona State University Dental Anthropology System (ASU-DAS) [40]; all individuals were pooled independent of age and sex as is practice in population studies [40–43]. Data were recorded for all teeth available but, as indicated by Turner and Scott’s individual count method [43, 44], when antimeres differed only the one with the highest degree of expression was scored. Breakpoints were selected for each of the dental traits

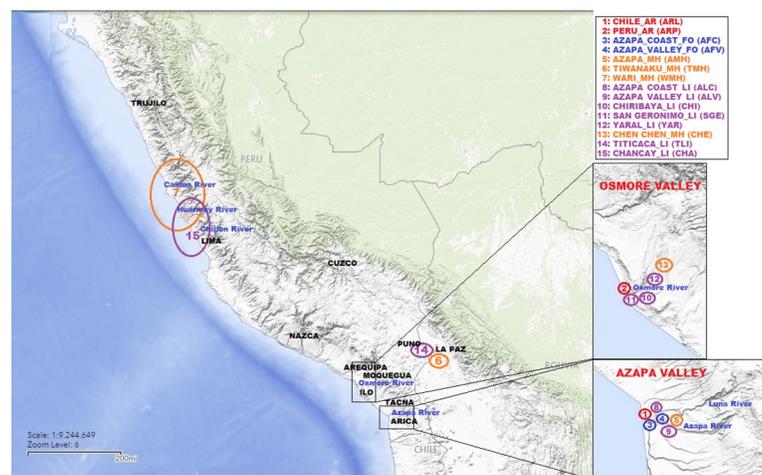


Fig 1. Location of the fifteen pre-Inca sites from Peru, Bolivia and northern Chile included in the study.

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Table 1. Sites analyzed.

	GROUP	ACRONYM	SAMPLE SIZE	PERIOD	CHRONOLOGY	REGION	DATA ORIGIN
1	CHILE_AR	ARL	84	Archaic	5000–1000 BC	Northern Chile	Sutter 1987
2	PERU_AR	ARP	16	Archaic	5000–1000 BC	Southern Peru	Sutter 1987
3	AZAPA_COAST_FO	AFC	62	Formative	1000 BC—200 AD	Northern Chile	Sutter 1987
4	AZAPA_VALLEY_FO	AFV	82	Formative	1000 BC—200 AD	Northern Chile	Sutter 1987
5	AZAPA_MH	AMH	10	Middle Horizon	300–700 AD	Northern Chile	Sutter 1987
6	TIWANAKU_MH	TMH	35	Middle Horizon	200–1100 AD	Southern Peru/Bolivia	Present study
7	WARI_MH	WMH	65	Middle Horizon	500–1100 AD	Central Coast Peru	Present study
8	AZAPA_COAST_LI	ALC	43	Late Intermediate	1000–1476 AD	Northern Chile	Sutter 1987
9	AZAPA_VALLEY_LI	ALV	205	Late Intermediate	950–1425 AD	Northern Chile	Sutter 1987
10	CHIRIBAYA_LI	CIR	189	Late Intermediate	900–1450 AD	Southern Peru	Sutter 1987
11	SAN_GERONIMO_LI	SGE	58	Late Intermediate	900–1450 AD	Southern Peru	Sutter 1987
12	YARAL_LI	YAR	52	Late Intermediate	900–1450 AD	Southern Peru	Sutter 1987
13	CHEN CHEN_MH	CHE	119	Middle Horizon	600–1000 AD	Southern Peru	Present study
14	TITICACA_LI	TLI	199	Late Intermediate	900–1450 AD	Southern Peru/Bolivia	Present study
15	CHANCAI_LI	CHA	372	Late Intermediate	900–1450 AD	Central Coast Peru	Present study
	TOTAL		1591				

List of sites analyzed, acronyms, chronology and geographic location. The Tiwanaku_MH group includes 4 individuals from Huaqui and 31 indicated in the museum repositories as Tiwanaku affiliated Middle Horizon samples from Bolivia or Peru; the Wari_MH group includes 2 individuals from Chimucapac, 2 from Huarney and the 61 samples from Pachacamac that were attributed in the museum repositories to the Middle Horizon.

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considered and the original data were simplified into a presence/absence dichotomy that was used to calculate trait frequencies for each of the 15 groups considered in the study (S17 Table).

In order to ensure that interobserver error would not compromise results, the authors responsible for scoring data calibrated with one another reaching agreement of more than 90% for each trait [45]. In order to ensure comparisons could be conducted with the published dataset, trait by trait comparisons were conducted on a sample from Chiribaya (37 maxillas and 26 mandibles) that had been scored by both Sutter and Coppa discarding, as indicated by Nichol and Turner [46], all traits that showed discrepancies in Net Mean Grade Difference greater than 20%.

Forty-seven traits were initially scored and considered suitable, on the basis of interobserver concordance indications [46] for the present analysis. Eight of these were then discarded due to the extremely low variation between samples, reducing to 39 the number of traits used in this study (24 maxillary and 15 mandibular traits) for the 1591 individuals pooled into 15 groups on the basis of chronology and provenance (see S17 Table for trait frequencies and breakpoints).

Biological similarities between samples were assessed through maximum likelihood (ML) and the reliability of the results was appraised through bootstrap analysis. The results obtained were corroborated conducting principal component analysis (PCA), mean measure of divergence (MMD) with the Freeman-Tukey angular transformation for high (>0.95) or low (<0.05) trait frequencies [47, 48], multi-dimensional scaling (MDS), and cluster analysis (CA) by means of Ward's grouping method. Due to the fact that multivariate tests can produce slightly different results that depend on the specific grouping method and algorithm used [41], the present analyses are based on a selection of multiple quantitative methods so as to ensure that affinities observed between samples in more than one elaboration may be confidently considered the product of real similarities and not statistical artifact [20].

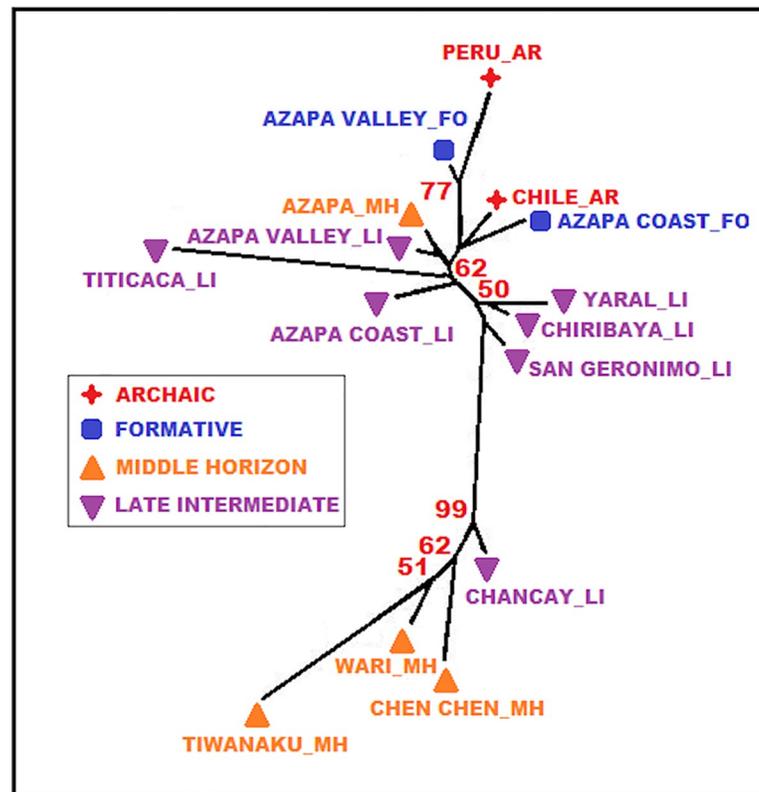


Fig 2. Maximum likelihood unrooted tree with bootstrap values. Node values are expressed out of 100 iterations.

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Results

The ML analysis (Fig 2) divides the samples into two separate and very distinct clusters positioned at the opposite ends of an unrooted tree with a bootstrap value of 99 that strongly supports this first separation of the samples. The first of these two clusters includes the three Peruvian Tiwanaku and Wari affiliated Middle Horizon samples (Tiwanaku_MH, Wari_MH and Chen Chen_MH) as well as the Late Intermediate sample from Chancay.

The second cluster includes both of the Archaic groups (Peru_AR and Chile_AR), all the Azapa Valley ones (coastal and inland), the Late Intermediate groups from the Osmore Valley (Chiribaya_LI, Yaral_LI and San Geronimo_LI) and the Late Intermediate sample from Titicaca_LI. A more detailed observation of the first of the two main clusters indicates that the three Middle Horizon samples show greater similarities to one another than they do to the Late Intermediate sample from Chancay_LI that branches onto the cluster last. Furthermore, within the second, larger cluster all of the more ancient groups form a sub-cluster, showing greater similarities one to the other than they do to the later Middle Horizon and Late Intermediate groups. Lastly, the Late Intermediate sample from Titicaca is characterized by a particularly long branch that somewhat sets it apart from the other samples.

To corroborate the results obtained through the ML statistic, we conducted a cluster analysis applying Ward's grouping method (Fig 3) and obtained a similar separation of the samples: a first cluster with the three Middle Horizon Peruvian samples and the Late Intermediate one from Chancay and a second with all of the remaining samples.

Once again, this second, larger cluster merits some further consideration. It shows that the Late Intermediate sample from Titicaca is the one that diversifies the most from the other

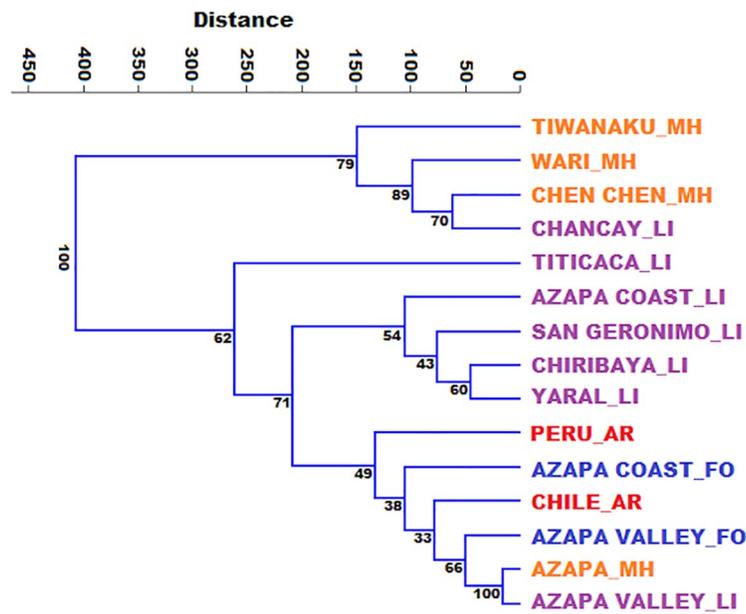


Fig 3. Cluster analysis based on Ward's grouping method. Node values are those obtained through 100 iterations.

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samples of the cluster for, in 62% of the iterations, it is placed on an independent branch. Furthermore, in 71% of the iterations, the remaining samples separate into two further clusters: one containing most of the Late Intermediate samples and the other containing most of the earlier ones.

When applying principal component analysis (PCA) (Fig 4), the three Middle Horizon Peruvian groups, together with the Late Intermediate Peruvian sample of Chancay_LI, plot

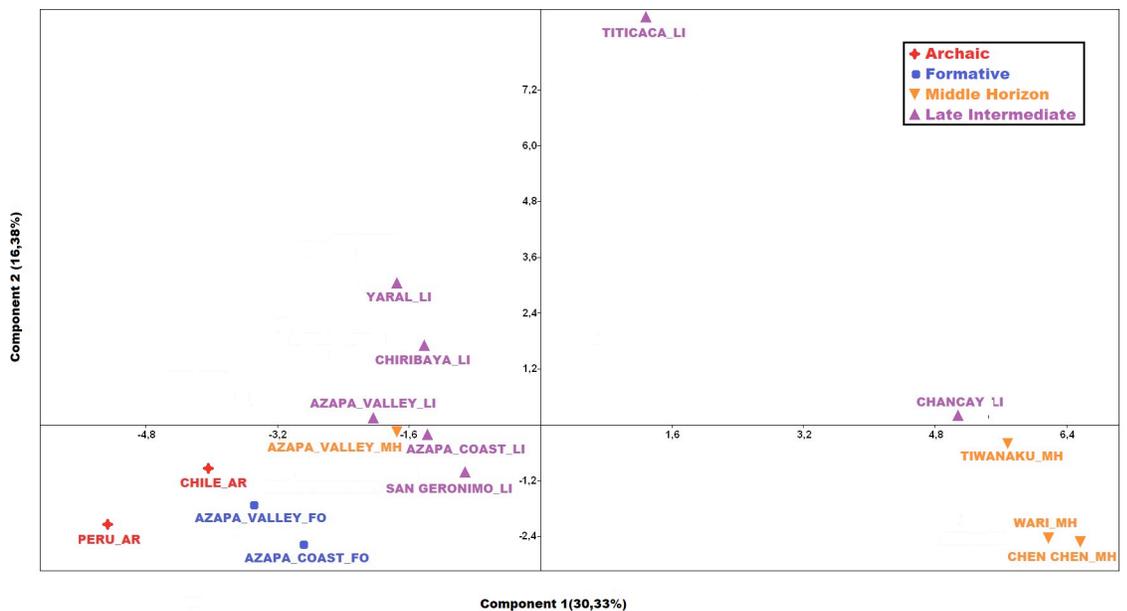


Fig 4. Bi-dimensional scatterplot from principal component analysis.

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along the positive axis of the first component (which explains 30.3% of total variance) distancing themselves from all of the others. The remaining groups plot along the negative axis of the first component with the sole exception of the Late Intermediate sample from Titicaca_LI which plots at values intermediate between these two main groups of samples. Apart from this first, clear cut, separation, the first component separates the samples positioned along its negative axis into three separate sub-groups according to a chronological gradient: the first with the two more ancient Archaic groups, the second with the two Formative ones, and the last with all of the later Middle Horizon and Late Intermediate groups. The second component (which explains 16.4% of variance) separates the Late Intermediate sample from Titicaca from all of the others. It, likewise, separates somewhat samples based on chronology, positioning the more ancient ones in the direction of the more negative values. Correlation coefficients that explain this distribution and the influence of each trait along each of the first two axes are listed in [Table 2](#) (in bold those with correlation coefficients greater than 0.7).

Distance between groups was assessed through Mean Measure of Divergence (MMD) ([Table 3](#)). The results obtained support the previously observed distributions and enable some further considerations. Almost all resulting inter-cluster distances are significant whereas almost all intra-cluster ones are not (and even when they are significant, they are, always, short). In particular, the MMD statistic indicates that the three Middle Horizon Wari and Tiwanaku affiliated sites (Tiwanaku_MH, Wari_MH and Chen Chen_MH) are extremely similar one to the other and to the later Late Intermediate sample from Chancay (the only significant distance the statistic is capable of detecting is a short one of 0.06 between the samples of Chen Chen_MH and Chancay_LI). A similar situation characterizes the smaller of the two sub-clusters from the Azapa and Osmore valleys given that none of the distances separating the Archaic or Formative samples are significant. Though always maintaining distances that are inferior to those shown with samples from the first cluster, the later samples of the second cluster start to show some significant distances both between one another and with the samples of the earlier Archaic and Formative periods of the other sub-cluster.

The Late Intermediate sample from Titicaca shows significant distances to all other samples considered and, in particular, to the Middle Horizon one from Chen Chen.

This analysis consequently supports the reliability of the previously observed clusters and, likewise, enables some further insight into the relations that might exist between the samples of the larger cluster.

Discussion

The last decades have witnessed an intensification of bioarchaeological investigations on the ethnicity, population dynamics, cultural and genetic changes that characterized the Osmore [7, 17–21, 23, 25, 28, 49] and the Azapa [2, 15, 22, 24, 26] valleys in pre-Inca times. Such studies have, in particular, focused on determining the extent of population movements along the valleys or in-between valleys; on establishing if the different, culturally distinct, communities that inhabited these valleys were the product of in situ microevolution or if, instead, they arrived through migrations; on finding the ties between these supposed migrant populations and the autochthonous ones from the same regions, and on understanding the manner in which the great polities that arose during the Middle Horizon spread their hegemony. However, even though such studies have, undoubtably, given important contributions, much is yet to be understood.

The present study indicates that even though some differences might be detected within the samples from the Azapa and Osmore valleys, these are, when viewed in a wider regional perspective, quite similar one to the other, especially in the earlier phases. All samples from these

Table 2. Principal component analysis factor loadings.

Trait	Component 1		Component 2	
I ¹ Shoveling	0.768 *		-0.1695	
I ² Shoveling	0.7493 *		-0.3567	
I ¹ Double Shoveling	0.4047		-0.4495	
I ² Double Shoveling	0.7314 *		-0.3187	
I ² Interr. Groove	0.6384		0.4898	
I ² Tuberculum Dentale	0.9089 *		0.1865	
C ¹ Distal Accesory Ridge	0.5264		-0.2467	
P ³ Cusp Number	0.1387		0.8686 *	
P ⁴ Cusp Number	-0.2246		-0.05529	
M ¹ Metacone	0.4095		0.181	
M ² Metacone	0.4294		0.1963	
M ³ Metacone	-0.0112		-0.1174	
M ² Hypocone	-0.05323		0.1233	
M ³ Hypocone	0.6264		0.1364	
M ¹ Cusp 5	-0.5799		0.06724	
M ² Cusp 5	-0.6901		0.01478	
M ³ Cusp 5	0.3084		0.8341 *	
M ¹ Carabelli's Cusp	0.4839		0.5774	
M ² Carabelli's Cusp	0.3661		0.3089	
M ³ Carabelli's Cusp	0.06234		0.746 *	
M ¹ Parastyle	0.3489		0.2539	
M ² Parastyle	0.7407 *		-0.03857	
M ³ Parastyle	0.6144		0.07881	
M ¹ Enamel Extencion	-0.2955		0.1643	
M ² Enamel Extencion	-0.3455		0.6517	
M ³ Enamel Extencion	0.2742		0.3635	
P ³ Root Number	0.2903		-0.4369	
P ⁴ Root Number	-0.474		-0.03688	
M ³ Peg Shaped	-0.6997		-0.3038	
I ² Peg Shaped	0.5339		-0.4488	
M ³ Cong. Absence	0.1514		-0.1539	
I ₂ Shoveling	0.7487 *		-0.1588	
M ₂ Groove Pattern	-0.505		-0.613	
M ₃ Groove Pattern	-0.3386		-0.3911	
M ₁ Cusp Number	-0.8767 *		0.308	
M ₂ Cusp Number	0.9164 *		-0.1936	
M ₁ Protostylid	0.4364		0.6467	
M ₂ Protostylid	0.3741		0.712 *	
M ₃ Protostylid	0.5503		0.5818	
M ₁ Cusp 5	-0.57		0.3649	
M ₂ Cusp 5	-0.9202 *		0.1567	
M ₃ Cusp 5	-0.8411 *		0.09038	
M ₁ Cusp 6	-0.8049 *		0.3696	
M ₂ Cusp 6	-0.7602 *		0.4264	
M ₃ Cusp 6	-0.5205		0.613	
M ₁ Cusp 7	-0.3436		0.3209	
M ₂ Cusp 7	-0.3238		0.2238	

(Continued)

Table 2. (Continued)

Trait	Component 1		Component 2	
M ₃ Cusp 7	-0.2473		0.03094	
M ₁ Root Number	0.2098		0.8169 *	
M ₂ Root Number	0.5587		0.4633	
M ₂ Root Number	0.8299 *		0.04105	
M ₃ Cong. Absence	-0.04553		-0.3845	
Variance explained				
by the components	15.7728		8.5184	
Percentage of variance explained				
by the components	30.33		16.38	
Total percentage of variance				
explained by the first two components		46.71		

Principal component analysis factor loadings. Correlation coefficients greater than 0.7 are in bold and are followed by an asterisk.

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valleys, with the sole exception of the supposed Tiwanaku outpost of Chen Chen, cluster showing little or no differentiation. This indicates the presence of a shared gene pool, interactions or continuity from at least the Archaic to the Formative. The proximity in all analyses of the Late Intermediate valley samples to the earlier Archaic and Formative ones from the same areas suggests that this might, in most cases, be true beyond the Formative and that there might have been continuity throughout the period taken into consideration. The data indicate, however, that there was at least one notable exception to this represented by the Middle Horizon sample of Chen Chen (Chen Chen_MH) from the Osmore Valley that positions itself, in all analyses, closest to the Middle Horizon Wari and Tiwanaku affiliated groups than it does to the other samples from the Osmore Valley. This and the great difference observed, in all statistics, between this sample and the other Middle Horizon one from the relatively nearby Azapa

Table 3. Mean measure of divergence matrix.

	ARL	ARP	AFC	AFV	AMH	TMH	WMH	ALC	ALV	CIR	SGE	YAR	CHE	TLI	CHA
ARL		0.035	0.019	0.015	0.009	0.058	0.045	0.020	0.009	0.010	0.016	0.014	0.016	0.018	0.026
ARP	-0.067		0.040	0.036	0.031	0.076	0.061	0.041	0.031	0.031	0.036	0.035	0.036	0.039	0.044
AFC	0.005	-0.043		0.020	0.014	0.062	0.047	0.026	0.014	0.015	0.020	0.018	0.020	0.023	0.030
AFV	0.003	-0.072	0.017		0.010	0.058	0.044	0.021	0.010	0.011	0.016	0.014	0.016	0.019	0.026
AMH	0.054	-0.041	0.041	0.023		0.053	0.040	0.015	0.004	0.005	0.011	0.009	0.011	0.013	0.021
TMH	0.273	0.151	0.359	0.249	0.230		0.087	0.064	0.053	0.054	0.059	0.057	0.059	0.061	0.069
WMH	0.290	0.221	0.320	0.311	0.240	-0.079		0.050	0.039	0.040	0.046	0.043	0.046	0.046	0.057
ALC	0.014	-0.080	0.007	0.020	0.013	0.190	0.175		0.015	0.016	0.022	0.020	0.022	0.024	0.032
ALV	0.037	-0.038	0.023	0.012	-0.005	0.237	0.251	0.025		0.005	0.011	0.008	0.011	0.013	0.021
CIR	0.078	0.015	0.142	0.075	0.095	0.139	0.144	0.031	0.084		0.011	0.009	0.011	0.013	0.022
SGE	0.037	-0.023	0.074	0.043	0.066	0.164	0.131	0.009	0.055	0.032		0.015	0.017	0.019	0.028
YAR	0.107	0.037	0.184	0.111	0.097	0.198	0.230	0.067	0.098	0.020	0.039		0.015	0.017	0.025
CHE	0.484	0.401	0.511	0.469	0.453	0.022	0.020	0.415	0.445	0.322	0.284	0.382		0.019	0.027
TLI	0.270	0.206	0.331	0.331	0.270	0.259	0.372	0.264	0.262	0.330	0.305	0.363	0.661		0.028
CHA	0.273	0.158	0.266	0.273	0.200	-0.032	-0.077	0.186	0.213	0.177	0.141	0.235	0.063	0.305	

Mean measure of divergence matrix. The values of divergence values are listed in the lower left portion, the corresponding standard deviations in the upper right one (bold values indicate statistically significant distances). Group acronyms are listed in Table 1.

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Valley, suggests that the population movements that characterized the Middle Horizon might have influenced to various degrees the inhabitants of the valleys perhaps destabilizing to some extent, and for a limited period of time, the morphological homogeneity of the populations inhabiting them.

Overall, the Middle Horizon saw great changes that must be taken into account in the interpretation of the results obtained. Owen [50] suggested that the collapse of the Tiwanaku state at the end of the Middle Horizon period led to a second diaspora of Tiwanaku colonists towards the coast, a continuity that found support in other studies [7, 25, 51–53]. In contrast, Lozada Cerna [21, 49] argued against this hypothesis suggesting that the Late Intermediate Chiribaya groups did not have particular ties to the Middle Horizon Tiwanaku colonists of the Osmore Valley. In support of Lozada Cerna's interpretation, Sutter [2, 24] found morphological differences between the coastal and the inland sites of the Azapa Valley and noted that they all separated from the Late Intermediate Chiribaya, San Geronimo and Yaral groups from the coastal Osmore Valley. The differences observed with the Chiribaya groups led him to interpret the Osmore Valley coastal drainage Chiribayas as a likely result of the Late Intermediate Tiwanaku diaspora [51].

Biocultural markers such as cranial deformation patterns have suggested the presence of strong ties between the Tiwanaku highland population and the one from their enclaves in the Middle and Upper Osmore Valley (i.e., Chen Chen) [18, 19]. Even though supported by archaeological material culture [3] and isotopic evidence—strontium and oxygen that indicate migration into the Osmore Valley of individuals from regions characterized by isotopic values compatible with those of the southern Titicaca basin [29, 54]—these ties do not preclude the existence of relations between Chen Chen and some of the nearby Wari affiliated outposts of the Middle Osmore Valley, such as Cerro Baul. Evidence of this was found by us in a recent study conducted on a restricted portion of the samples used in the present analysis [20], in which we detected affinities between the Middle Horizon site of Chen Chen and the Wari affiliated Middle Horizon sample from Pachacamac. The relations detected among samples suggested these ties had been anything but negligible and that, had isolation by cultural barriers occurred [55], as proposed by Goldstein [3], Chen Chen should have differed more from those groups. Furthermore, the results suggested that either these groups derived from a common gene pool or that perhaps, as also hypothesized by Moseley et al. [30], after the establishment of Cerro Baul and of the other outposts, Wari colonists came into direct contact with their Tiwanaku counterparts.

The inclusion, in the present study, of the Tiwanaku affiliated sample from the southern Titicaca basin provides further support to the hypotheses according to which Wari and Tiwanaku affiliated colonists shared a common gene pool or came into contact in the Middle Osmore Valley. In all the multivariate analyses conducted the three Middle Horizon Wari and Tiwanaku affiliated samples (Chen Chen_MH, Tiwanaku_MH and Wari_MH) cluster or plot close to one another, far from the Late Intermediate sample from Titicaca.

Overall, our results suggest that the Osmore and Azapa drainages shared a common gene pool from archaic times to the Formative period and that this period was followed by one—the Middle Horizon—characterized by substantial changes, population movements, cosmopolitanism and the establishment of Wari and Tiwanaku outposts [3, 53]. They, furthermore indicate that, these migrations impacted substantially the population structure of the Middle Horizon groups considered but that they did so in a transient manner given that the later (Late Intermediate) samples of the region plot, in all analyses, in the vicinities of their Archaic and Formative predecessors. These results consequently fit well with those obtained through aDNA that, likewise, indicated population continuity in the region [53].

The distances observed between the Late Intermediate groups from Chiribaya, San Geronimo and Yaral combined to those seen between the Middle Horizon Tiwanaku affiliated sites

of Tiwanaku_MH and Chen Chen_MH fit Lozada Cerna's theory [21, 49], according to which the Late Intermediate coastal sites in the Osmore drainage were independent from the Tiwanaku colonies, better than they do with Owen's [51] hypothesis of a second Tiwanaku diaspora. However, we must maintain caution and view the results as the indicators they are and not as unequivocal proof given that the Tiwanaku affiliated sample analyzed in the present study dates to the Middle Horizon, while the ones from Yaral, San Geronimo and Chiribaya date to the Late Intermediate. Moreover, the supposed second diaspora would have occurred at the onset of the Late Intermediate period, just after the fall of the Tiwanaku state around AD 1000–1100 [10, 56–58], as a likely consequence of climatic changes—or more specifically drought—and the sociopolitical collapse in the *altiplano* [11, 59–62].

Furthermore, the diaspora could have given rise to low-status groups that integrated, as minorities, in the coastal Osmore Valley populations [50] or, given the ethnic diversity attributed to the Tiwanaku population,—described as characterized by independent “moieties, ethnic groups, or maximal ayllus” [3: p51]—we could simply have included Tiwanaku samples that are not representative of those that supposedly migrated towards the coast.

The three Middle Horizon, Tiwanaku and Wari affiliated samples cluster even though distant both geographically and culturally; consequently, depicting a different side of the population history of the region. There is little doubt that Tiwanaku and Wari represented two separate and independent polities, as indicated by the archaeological evidence [3]. Both of their outposts in the Osmore Valley produced ceramics, similar in style to those found in their respective heartlands, using locally available clay procured within their respective territories [63]. Even though the respective inhabitants of the enclaves of these two polities certainly imported cultural practices from their respective homelands, the present study reveals patterns of mobility and population dynamics within southern Peru and northern Chile that contrast with the generalized ideas of separation based on cultural identity for these two Middle Horizon polities.

The proximity shown in all analyses between the three Middle Horizon Wari and Tiwanaku affiliated groups indicates that while these two groups can easily be separated on the basis of culture, the similarities observed in dental morphology suggest that there may have been significant movement of peoples that kept a generally widespread homogeneity between these Andean populations making them much harder to distinguish one from the other.

The patterns of population dynamics in Central-Southern Peru are also discussed by Knudson and Tung [35: p.307], who state that the presence of “non-local individuals at the site of Beringa derived from geologic zones located in the Pacific coastal areas, of what is now southern Peru, parts of Bolivia, or northern Chile, is consistent with the notion that the Middle Horizon was marked by expansive trade networks and population movement, likely influenced by local, historically durable trade practices and policies of the Wari empire”. Population movement was, for example, detected in a number of studies among which one conducted on trophy heads from the Wari site of Conchopata using strontium isotopes that shows that trophy heads are non-local therefore suggesting that the Wari military agents moved between geographic regions taking back captives or at least their remains [64]. The recent ancient DNA studies conducted in the region [53, 65] likewise find evidence of genetic exchanges with other regions as well as evidence of long-standing population continuity. A similar pattern of genetic homogeneity finds support in Kemp et al. [34], who suggest genetic continuity between Wari and post-Wari populations based on mtDNA analysis. Nonetheless, they do not rule out that the genetic continuity could be the result of an overall homogeneity in the region, which may have masked possible female migrations into Wari territory after its collapse. Similarly, Lewis et al. [66] find that mtDNA changes between Chen Chen and a large number of ancient and extant South American populations indicates the presence of strong haplogroup homogeneity

in the region. They assess that this homogeneity existed well before the Middle Horizon; i.e., well before the period that witnessed the rise of both the Wari and the Tiwanaku polities. According to Lewis [67], the mtDNA changes, detected between the ancient and the contemporary groups in the region, are more likely the result of genetic drift than of genetic exchange or migrations through time.

Interesting are likewise the distributions shown by the Late Intermediate samples of Chancay_LI and Titicaca_LI. The first of the two plots with the Middle Horizon Wari and Tiwanaku affiliated samples in all statistical analyses, which could be the result of geographic continuity in central Peru from the Middle Horizon Wari affiliated group to the Late Intermediate one from Chancay. Conversely, the Late Intermediate sample from Titicaca plots in the opposite cluster from the earlier Middle Horizon Tiwanaku affiliated group in its geographic vicinity. This group distances itself from all the groups considered in the present study in a position that does not appear to fit properly with any of the proposed models. Its positioning does however seem to find explanation when analyzed in light of genetic drift, isolation, and founder effect concepts. The Titicaca collection comes from the highlands along the northwestern side of the Lake Titicaca, at the opposite end of the Tiwanaku State and far away from the sphere of influence of the Wari Empire. The distance shown between this group and all the other—geographically closer—groups in southern Peru, could consequently be the result of isolation by distance and genetic drift.

The results obtained outline an important trend suggesting that, in general, population structure in the area under investigation was shaped by two contrasting phenomena: on one side population continuity, that characterized both the earlier and the later phases and, on the other, change such as the one detected during the Middle Horizon in consequence of the rise and expansion of the Tiwanaku. Such results are perfectly aligned to all those indicating that geographic proximity and geographical barriers are powerful forces capable of shaping populations and their genetic assets [68–70] especially prior to the appearance of large-scale population movements and the rise of strong polities or states capable of extending their hegemony over vast areas. This is due to the fact that, once technological advances and cultural changes brought to large scale phenomena—such as, for example in Europe, the spread of the Neolithic [42, 71–74], of the Bell Beaker [75], of the steppe related ancestry [76, 77] or of the Roman Empire [78]—the patterns became more complex. In such instances, one may often observe, as is the case in the present study, that temporal affiliation likewise contributes, at times notably, to the genetic structure of the populations considered, even across vast geographical space.

Supporting information

S1 Table. Collections, acronyms, chronologies and locations. List of collections analyzed, acronyms, chronological period and location.
(XLSX)

S2 Table. Chile_Archaic. Individual morphological data in the Chile_Archaic group.
(XLSX)

S3 Table. Peru_Archaic. Individual morphological data in the Peru_Archaic group.
(XLSX)

S4 Table. Azapa_Coast_Formative. Individual morphological data in the Azapa_Coast_Formative group.
(XLSX)

S5 Table. Azapa_Valley_Formative. Individual morphological data in the Azapa_Valley_Formative group.

(XLSX)

S6 Table. Azapa_Middle_Horizon. Individual morphological data in the Azapa_Middle_Horizon group.

(XLSX)

S7 Table. Tiwanaku_Middle_Horizon. Individual morphological data in the Tiwanaku_Middle_Horizon group.

(XLSX)

S8 Table.

(XLSX)

S9 Table. Azapa_Coast_Late_Intermediate. Individual morphological data in the Azapa_Coast_Late_Intermediate group.

(XLSX)

S10 Table. Azapa_Valley_Late_Intermediate. Individual morphological data in the Azapa_Valley_Late_Intermediate group.

(XLSX)

S11 Table. Chiribaya_Late_Intermediate. Individual morphological data in the Chiribaya_Late_Intermediate group.

(XLSX)

S12 Table. San_Geronimo_Late_Intermediate. Individual morphological data in the San_Geronimo_Late_Intermediate group.

(XLSX)

S13 Table. Yaral_Late_Intermediate. Individual morphological data in the Yaral_Late_Intermediate group.

(XLSX)

S14 Table. Chen_Chen_Late_Intermediate. Individual morphological data in the Chen_Chen_Late_Intermediate group.

(XLSX)

S15 Table. Titicaca_Late_Intermediate. Individual morphological data in the Titicaca_Late_Intermediate group.

(XLSX)

S16 Table. Chancay_Late_Intermediate. Individual morphological data in the Chancay_Late_Intermediate group.

(XLSX)

S17 Table. Frequencies. Frequencies of the 39 traits used for statistical analyses in the 15 groups according to their breakpoints.

(XLSX)

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