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East-West cranial differentiation in pre-Columbian populations from Central and North America

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Abstract

In a recent study we found that crania from South Amerindian populations on each side of the Andes differ significantly in terms of craniofacial shape. Western populations formed one morphological group, distributed continuously over 14,000 km from the Fuegian archipelago (southern Chile) to the Zulia region (northwestern Venezuela). Easterners formed another group, distributed from the Atlantic Coast up to the eastern foothills of the Andes. This differentiation is further supported by several genetic studies, and indirectly by ecological and archaeological studies. Some authors suggest that this dual biological pattern is consistent with differential rates of gene flow and genetic drift operating on both sides of the Cordillera due to historical reasons. Here we show that such East-West patterning is also observable in North America. We suggest that the "ecological zones model" proposed by Dixon, explaining the spread of the early Americans along a Pacific dispersal corridor, combined with the evolution of different population dynamics in both regions, is the most parsimonious mechanism to explain the observed patterns of within- and between-group craniofacial variability.
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Introduction

Recently, Pucciarelli et al. (2006a) found that South American highland populations situated on the Pacific side of the continent and the lowland groups occupying the eastern plains from Venezuela to Argentina, presented remarkable differences in terms of within- and between-group cranial morphology. This cranial differentiation is congruent with previously reported mtDNA differentiation patterns (Fuselli et al., 2003).

In the case of South America, the East-West differentiation has been putatively explained by geneticists and molecular biologists as the result of local processes of differential genetic drift and flow (Simoni et al., 2000; Tarazona-Santos et al., 2001; Llop et al., 2002; Fuselli et al., 2003). This is a highly attractive hypothesis since demography, social organization, and technological development were very different on each side of the Andes due to geographical and ecological conditions (Carneiro, 1970; Focacci and Erices, 1973; Meggers, 1979; Muñoz and Focacci, 1983; Bruhns, 1994). It is possible, however, that earlier processes such as the dispersal of initial groups along ecological corridors may be responsible for part of the current patterns of variation. Whatever the case, Amerindian

heterogeneity is the product of complex historic and structural processes that need to be more thoroughly studied in order to reconstruct the continent's early settlement and in situ population dynamics.

High levels of phenotypic diversity in the New World were recognized early on by researchers like Neumann (1942, 1952), who centered his studies on the analyses of the American Indians. Bass (1964) was also concerned with levels of variation between prehistoric Plains Indians. Although their morphometric work was heavily influenced by the typological approach that dominated physical anthropology during the first half of the twentieth century, they were well aware of the complicated influences that migration, group fission, and adaptive and plastic developmental responses to cultural influences such as dietary change and activity pattern could have on the skeleton.

The seminal work of Howells (1973, 1989) revealed and evaluated Amerindian heterogeneity using a perspective firmly grounded in population genetics, and taking advantage of the expansion of computer analysis and multivariate statistics. Howells demonstrated that cranial variation in Native Americans is surprisingly high given the area and the time depth of occupation (Howells, 1989), stating that "intraregional heterogeneity is greatest in Polynesia and the Americas, the two regions we can certify as the latest to be occupied. This goes counter to any expectation that such recency would be expressed in cranial homogeneity" (Howells, 1989: 83).

Formal settlement models like the one proposed by Greenberg et al. (1986) also dealt with Amerindian morphological variation, but focused on dental rather than craniofacial morphology. These authors support a pattern of North-South Amerindian linguistic, serological, and dental differentiation that could be viewed as the traces of three different waves of settlers (the so-called Amerindian, Na Dene, and Eskimos) coming from Asia at three different time periods.

Further research on North Amerindian heterogeneity can be found in Jantz et al. (1992), who re-studied the Boas anthropometric data and found strict patterns of geographic variation. Recently, and departing from Brace's data bank, Nelson (1998) specifically examined North American craniometric variation among Amerindian samples, and further compared their results with Neumann's (1952) assessment of regional variation (Nelson, 2006).

Some papers by Brace and Hunt (1990), Powell and Neves (1999), Brace et al. (2001, 2004), Jantz and Owsley (2001), and González-José et al. (2003) are concerned with Amerindian variation on a global, continental scale. In some cases, these analyses adopt a diachronic perspective and investigate the relationship among early-late Pleistocene through early Holocene Old and New World samples and modern Amerindians. These comparative analyses make the important observation that many early remains tend to fall outside the range of variation of most (but not all) modern East Asians and Amerindians, reflecting important morphological shifts occurring simultaneously with the early settlement of the Americas.

Modern genetic and phenotypic patterns of variation in North America clearly result from complex microevolutionary

historic and structural events, which may have acted simultaneously and were not necessarily mutually exclusive. Therefore, interpretation of modern patterns of craniofacial variation must take into account the putative influence of all the factors involved. Previous work focusing on South American groups can be of great help as a starting point to test for disparate evolutionary scenarios. Thus, in this work, we investigate whether the East-West craniofacial pattern of differentiation previously observed in South America (Fuselli et al., 2003; Pucciarelli et al., 2006a) can also be detected in Central and North America. We use a set of statistical analyses based on population genetic principles, in order to further investigate the possible role of structural aspects (such as differential levels of gene flow, gene drift, and spatial structuring), as well as historical ones (like the use of a Pacific dispersal corridor), as explanations for the current patterns of observed variation. This scenario can be tested by analyzing patterns of within- and between-group variation, and by using Paleoamerican, Westerner, and Easterner samples. We focus on these questions by testing the central hypothesis:

If a North-South expansion of the ancestral group of Beringians was later divided into two sub waves, one going through a Pacific corridor (ancestral to modern Westerners) and the other through the Atlantic plains (resulting in modern Easterners), assuming initially similar population sizes and a constant rate of North-South displacement, then the pattern of phenotypic differentiation will be characterized primarily by East-West differences, rather than differences consistent with pure isolation-by-distance.

Furthermore, if each putative lineage did not evolve within the same microevolutionary landscape due to differences in population density on either side (e.g., low density in the wide plains of the East, relative to higher density in the narrow corridor of the West), then genetic drift would have been a primary shaper of diversity in Easterners, while gene flow would have played an important role among Westerners. Therefore, an East-West differentiation is expected in terms of patterns of within- and between-group variation. This scenario can be tested by comparing within- and between-group variation in western and eastern sets of samples, as well as examining the spatial patterning of craniofacial differences in both regions.

Two subsidiary hypotheses will also be considered. The first addresses whether a complementary North-South differentiation process occurred within each region (East and West):

If a North-South expansion of people was coming from the Bering Strait to the Fuegian archipelago, then a North-South differentiation gradient would be expected.

If this complex settlement and dispersal scenario is supported by the data, then the New World variability should be explained not just by the classical mechanisms based on number of waves from Asia, variability in the ancestral Asian population, etc., but also taking into account the particular dynamic of dispersions across the continent. Moreover, the acceptance of this complex scenario would mean that the pattern of East-West differentiation found in South America is indicative of a more widespread phenomenon that spanned almost the entire continent and, thus, may result from a single process

linked to differential rates of genetic drift/gene flow. The second subsidiary hypothesis relates to the geographic position of the Paleoamericans before the putative East-West split:

Assuming Paleoamericans were responsible for the first expansion across the continent, and considering that they were the ancestor of both putative population groups, then an intermediate distribution of the Paleoamericans with respect to Easterners and Westerners would be expected.

Material and methods

The samples

Three-hundred and seventy-five non-deformed male skulls from the late Holocene (1200–1400 AD) were examined, derived from 27 regions representing well-recognized groups of late pre-Columbian American Indians (Table 1). Given the small size of some samples, we averaged the jackknifed

classification matrix from the discriminant analysis. The result was 87.5% of correct placements. Samples were further classified as Westerners (W) when they inhabited regions along the Pacific corridor, extending from the West Coast to the Rocky Mountains, and as Easterners (E) when they were found in the area extending from the eastern border of the Rocky Mountains chain to the Atlantic Coast.

In regions where an uninterrupted chain was absent or not evident, the boundary was set where small chains of mountains, sporadic elevations, compact fields of trees and shrubs, or any other geographical feature allowed us to differentiate a long and narrow Pacific strip from the wide Western plains. In Central America, for instance, samples from the Atlantic Coast (Honduras) and the Antillean Islands (Puerto Rico) were considered as Easterners, while the sample from the Pacific Coast (Costa Rica) was considered Westerners. In Mexico, the division was provided by an imaginary line parallel to the Pacific Coast passing through the Mexico Basin and

Table 1
Sample description

Sample	Code	n	Region	Location	Main ethnic group	Chronology
Artic Region						
01-Aleuts (Krauss, 1980)	Ale	33	W	Alaska	Aleut	1400 AD
02-Pacific Eskimos	Kon	12	W	Alaska	Koniag, Chugach	1400 AD
03-West Coast	Inu	12	E	Greenland	Inuit	1400 AD
Southeast Region (Florida)						
04-Safety Harbor	SHa	10	E	Pinellas	Tacobaga, Apalachee	1300 AD
05-Cape Cañaveral	CCa	13	E	Orange	Orange, Ais	1300 AD
06-Horr's Island (Sassman and Anderson, 2004)	HoI	13	E	Collier	Mounds, Calusa	1300 AD
07-Perico Island (Sassman and Anderson, 2004)	PII	14	E	Manatee	Uzita, Calusa	1300 AD
Southeast Region (Arkansas)						
08-Wichita-Caddo (Wedel, 1941)	WCa	13	E	Mississipi	Wichita, Caddo	1200 AD
North California						
09-Sacramento	Sac	31	W	Yolo-Sacramento	Patwin, Wintuan	1400 AD
10-Tulamniu	Tul	10	W	Kern	Yokut (Southern)	1400 AD
11-Centerville (Lilliard et al., 1939)	Cen	10	W	Alameda	Hokan, Penutian	1400 AD
South California						
12-La Patera	LPa	18	W	Santa Barbara	Coastal Chumash	1400 AD
13-San Juan Capistrano	SJC	11	W	Santa Barbara	Coastal Chumash	1400 AD
14-San Luis Obispo	SLO	11	W	San Luis Obispo	Coastal Chumash	1400 AD
15-Santa Rosa Island	SRI	10	W	Santa Barbara	Island Chumash	1400 AD
16-San Miguel Island	SMI	13	W	Santa Barbara	Island Chumash	1400 AD
17-Santa Cruz Island	SCI	22	W	Santa Barbara	Island Chumash	1400 AD
18-San Nicolas Island	SNI	10	W	Ventura	Island Gabrielino	1400 AD
Texas						
19-Comanche	Com	10	E	South Plains	Shoshon	1400 AD
Mexico						
20-Paleoamerican	Pal	5	P	Mexico Basin	Metro Balderas, etc.	11000 BC
21-Tlatelolco	Tle	10	W	Mexico Basin	Mexicas	1300 AD
22-Isla Angel de la Guarda (North, 1908)	IAG	11	W	Baja California	Kilíwa, Gimiel, etc.	1400 AD
23-Pericú	Pei	20	W	Baja California	Pericú	1400 AD
24-Chihuahua	Chi	10	W	Chihuahua St.	Tarahumara, Concho	1400 AD
25-Durango	Dur	10	W	Durango St.	Náhuatl, Huichol, etc.	1400 AD
Central America						
26-Honduras	Hon	10	E	Honduras	Copan (Maya)	1300 AD
27-Costa Rica	CRi	11	W	Costa Rica	Caribes, Borucas, etc.	1400 AD
28-Puerto Rico	PRi	12	E	Puerto Rico	Pre-Taino, Taino	1400 AD

coincident with the Sierra Madre. In California, the division was provided by several elevations, mainly the Sierra Nevada and the coastal range.

One sample of 11−9 ka old Paleoamericans (Pal) from the Mexico Basin was also included (Table 1). This was not used to evaluate East-West differentiation, but rather to estimate relative similarity of these ancient remains to either the Eastern or Western populations. Further details concerning the archaeological, linguistic, and ecological context of samples is available in Supplementary material.

Statistical procedure

All crania were measured by the craniofunctional method shown in Table 2 (Pucciarelli et al., 1990, 2003, 2006a,b; Dressino and Pucciarelli, 1999; Sardi et al., 2004; González-José et al., 2005). This method is based on the principles originally described by Klaauw (1948−52), as well as those of Moss and Young (1960), Moss (1961), and Moss's (1997a, b, c) Functional Matrix theory. Scaling and size effects were dropped out by simple and double z-standardizations, as follows: 1) z-standardization by rows, 2) z-standardization by columns (by transposing the data file), and 3) new transposition to restore the data file (Wilkinson et al., 2002). Normality of frequency distributions was tested by the one-sample K-S method.

Our analysis attempts to shed light on microevolutionary events which shaped differences within and among the western and eastern areas of North America, if indeed any differences exist. Also, we were concerned with inferring whether both regions developed similar population dynamics and spatial structure, and whether the pattern of variability observed in both sides is coincident with the one observed previously in South America. To address these issues, we performed four different analyses which examine patterns of among and within-group craniofacial variability at different hierarchical

Table 2
Variables used in this study

Number	Symbol	Name	Description	Caliper	Mode[*]
01	NL	neurocranial length	Nasion-Opisthocranium	Poech	projected
02	NW	neurocranial width	Eurion-Eurion	Spreading	direct
03	NH	neurocranial height	Basion-Vertex	Spreading	direct
04	FL	facial length	Inner Prosthion-Vomerobasilar	Poech	projected
05	FW	facial width	Zygion-Zygion	Spreading	direct
06	FH	facial height	Nasion-Prosthion	Poech	projected
07	ANL	anteroneural length	Glabella-Bregma	Poech	projected
08	ANW	anteroneural width	Pterion-Pterion	Spreading	direct
09	ANH	anteroneural height	Bregma-Vomerobasilar	Spreading	direct
10	MNL	midneural length	Bregma-Lambda	Poech	projected
11	MNW	midneural width	same as NW	Spreading	direct
12	MNH	midneural height	Basion-Bregma	Spreading	direct
13	PNL	posteroneural length	Opistion-Opisthocranium	Poech	projected
14	PNW	posteroneural width	Asterion-Asterion	Spreading	direct
15	PNH	posteroneural height	Lambda-Opistion	Poech	projected
16	OTL	otic length	timpanic bone posterior inferior end-midpoint of inner end of the petrous bone	Vernier	direct
17	OTW	otic width	external auditive width	Needle	direct
18	OTH	otic height	external auditive height	Needle	direct
19	OL	optic length	Dacrion-intersfenoidal foramen	Orbitometer	direct
20	OW	optic width	Dacrion-Ectoconquio	Poech	projected
21	OH	optic height	Mid Supraorbitary point-mid Infraorbitary point	Poech	projected
22	RL	respiratory length	anterior-posterior nasal spine	Spreading	direct
23	RW	respiratory width	maximum nasal width	Vernier	direct
24	RH	respiratory height	Nasion-Subnasal	Poech	projected
25	ML	masticatory length	lower border zygomatic synchondrosis-posterior border of the glenoid cavity	Vernier	direct
26	MW	masticatory width	anterior sulcus of the sphenotemporal crest-lower point of the zygotemporal synchondrosis	Needle	projected
27	MH	masticatory height	lower border of the zygotemporal synchondrosis-upper temporal line at the coronal intersection	Poech	projected
28	AL	alveolar length	external Prosthion-posterior alveolar border	Vernier	direct
29	AW	alveolar width	from left to right second-third molars width	Vernier	direct
30	AH	alveolar height	palatal deep at midsaggital/second-third molars	Palatometer	direct

[*] For the projected measurements, the skull must be placed laterally on a square 50×50 cm white cardboard for reaching an acceptable parallelism with the caliper bar and/or its branches. Positioning must be done by carefully rotating the skull up to an Auricular-Infraorbitary equalization (Frankfurt plane). Previously, the correct anterior-posterior and vertical placement of the skull must be ensured by the equalization of the Prosthion and Inion points with respect to the horizontal plane, and by the positioning of the palatal first molars perpendicularly to this plane. The Frankfurt orientation can be facilitated by a nylon thread placed not more than 1 cm above the skull, and held parallel to one of the cardboard lines. The thread must be taken away after the correct placement has been reached, and before measurement starts. Direct measurements may be made out of the Frankfurt orientation. It is recommended to take all projected measurements first and then all direct measurements, or vice versa.

levels of geographic clustering, ranging from analysis of separate local populations up to analyses of East-West groupings.

First, we computed a PCA analysis after the pooled within-group covariance matrix of the 24 standardized variables defining the minor components (Table 2). Due to its low sample size, the pooled within-group covariance matrix was obtained without considering the Paleoamerican specimens. The pooled within-group covariance matrix is used instead of the covariance one, in order to avoid bias in the computation of the principal components due to potential lack of proportionality between the *among population* and the *within population* covariance matrices. Computed in this way, the principal components not only represent axes of maximum covariance, but also the axes of least evolutionary resistance (Schluter, 1996; Marroig and Cheverud, 2005).

Information about distribution of individuals along the first PC's was represented in a two dimensional scatterplot of the PC scores. Dispersion of individuals corresponding to the three main geographical-chronological categories considered here (East, West, and Pal) was represented by ellipses of 90% distribution of each category.

Secondly, formation of meaningful sub groups among the samples was explored by computation of Mahalanobis distances calculated for all pairs of samples. The matrix of distances was graphically represented by means of a Multidimensional Scaling graphic (Manly, 1994). In order to avoid biased patterns caused by the presence of the Paleoamerican sample, we repeated the computation of distances excluding this sample from the analysis.

Third, we used the Relethford and Blangero (1990) model to explore patterns of within- and between-group variability among samples. The model states that when populations within a region exchange migrants with an outside source at equal frequency, the relationship between the average within-group variation and phenotypic distance to the centroid for each population should be linear. Deviations from the expected model will occur when one or more of the subpopulations have greater than average external gene flow. Consequently, those subpopulations that receive more extra-regional gene flow will have greater within-group variation than expected, while those receiving less than average external gene flow will have lower heterogeneity than expected. In this context, we computed levels of observed, expected, and residual variance for Easterners and Westerners (Harpending and Ward, 1982; Relethford and Blangero, 1990; Relethford and Harpending, 1994; Relethford, 1996). In addition, the Relethford and Blangero method was used to obtain the Fst parameter on different apportionments of samples: using the whole sample, only the Westerners, and only the Easterners. Fst was computed using the PC scores obtained as explained above, removing the Paleoamerican sample. To assess the impact of different assumptions about heritability (h^2), we replicated the computation of Fst's by using different values of h^2, including the widely accepted value of 0.55 for craniometric measurements. Thus, heritability was set to 0.25, 0.35, 0.45, 0.55, 0.65, 0.75, 0.85, and 0.95. Moreover, different demographic scenarios were evaluated considering different census sizes for the western
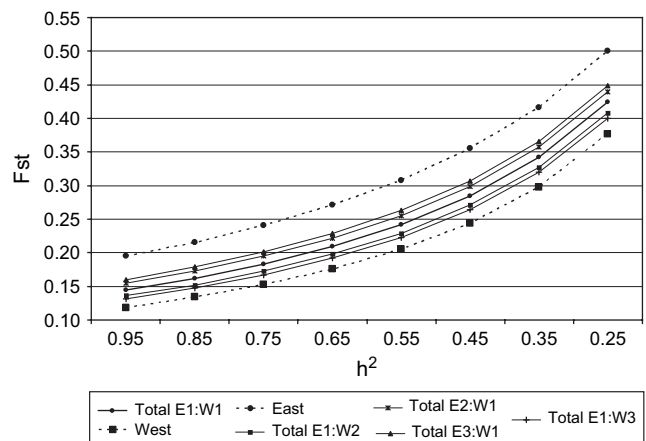

Fig. 1. Fst values for the total sample, Easterners, and Westerners, computed using different heritability values (horizontal axis). Different demographic scenarios are denoted as the relative census size for each region. Thus, E1:W1 denotes equal census sizes, E1:W3 denotes a Westerner census size three times larger than the Easterner, etc. Provided values are unbiased Fst, thus correcting the effect of sampling error due to small sample size as described by Relethford et al. (1997).

and eastern samples (see Fig. 1). Fst values obtained after different demographic circumstances and heritabilities cover a broad spectrum of putative scenarios, thus providing clearer insights about differences in population dynamics in both regions.

Further details concerning the theoretical and practical details of the Relethford and Blangero method can be found in Relethford and Blangero (1990), Relethford and Harpending (1994), Relethford (1996), and Relethford et al. (1997). Computations were made using the RMET 5.0 program, written by John Relethford.

Finally, spatial patterning of the whole modern sample was estimated by computing Mantel correlations (Mantel, 1967) among the matrix of craniofacial Mahalanobis distances and the matrix of geographic ones. According to the model of isolation by distance, if migration and gene drift are under balance among populations inhabiting a given region, then there will be a significant positive correlation between geographic and biological distances (Konigsberg, 1990). The geographical distance matrix reflects the distances in kilometers among archaeological sites from which the cranial samples were drawn, and biological distances correspond to the Mahalanobis distances matrix obtained above. As an additional test, we simulated a scenario in which Westerners and Easterners diverge into two population groups after entering the continent. To do so, we constructed a matrix of geographic distances in which any Westerner-Easterner pairwise distance is measured via a waypoint situated in central Alaska. The remaining Westerner-Westerner and Easterner-Easterner elements of the matrix are direct distances in km. Note that this matrix does not necessarily imply the absolute isolation after the settlement event, but reflects a lower probability of contact among populations situated in different regions. In addition, Smouse-Long-Sokal tests (Smouse et al., 1986) were used to yield partial matrix correlations. The Smouse-Long-Sokal

method extends Mantel's statistic to three or more matrices, and tests whether an association between matrix A and B is significant when one or more matrices (C, D, etc.) are held constant. The Smouse-Long-Sokal test was used to test partial correlations after removing the effects of geography as well as those of competing models. To evaluate if correlation and partial correlation coefficients are different from one another, the coefficients were jackknifed across populations, and then the differences among the replicated distribution of coefficients were tested.

The logical basis for the combination of the four procedures described above is grounded in the underlying principles of population genetics. For instance, if populations placed in the narrow western strip constituted a continuous genetic landscape where gene flow is the primary force shaping variability, as opposed to the eastern wide area where genetic drift could have been the force shaping variability, then we expect: a) a greater condensation of western individuals along the first PC's, b) minor levels of differentiation (Fst) in the West in relation to the East, and c) a positive and significant correlation among geographic and craniofacial distances, with the matrix simulating a Westerner and Easterner separation positively correlated with biological distances. Additionally, if the first dispersal across the continent occurred mainly along a Pacific corridor, then craniofacial distances among Paleoamericans and their supposed direct descendants (Westerners) should be lower than the distances between Paleoamericans and Easterners. Departures from these expected and straightforward patterns provide interesting clues about further mechanisms or historical events of potential importance during the evolution of North American human populations.

Results

The percentage of total variance explained by the first three principal components was 35%. The low percentage of variance explained reflects the fact that minor component variables are quite independent from on another. In comparison with classical measurements, where many measurements overlap and hence tend to contribute to the same PC, the low value of variance explained here is an expected result. The first PC was mainly explained by variation on the length dimension of the cranium, focused on the anterior and posterior neurocranium, (positive correlation with the first PC). Additionally, the length of the middle and the height of the posterior neurocranium, as well as the width of the masticatory component, contributed with high negative scores to the first PC. The second PC is driven mainly by variation in width dimensions of the anterior, middle, and posterior neurocranium, (negative correlation with the second PC), in addition to the important positive contribution of the length of the respiratory and alveolar components to this PC. Facial traits appear to be important on the third PC, which was mainly dominated by variation in respiratory length and width, and a contribution of orbital width. In conclusion, the principle component factors were differentially influenced by the craniofacial variables. While the first component primarily reflects variation

in neurocranial height, the second component reflects variation in neurocranial width, and the third reflects facial variation.

The scatterplot based on mean group scores along the first two components (Fig. 2a) shows that Westerners (except CRi) and Paleoamericans tend to occupy the positive values of the first PC and negative values of the second PC, whereas Easterners tend to be placed along the negative values of the first PC and positive values of the second one. A plot of individual scores from the same analysis with 90% distribution ellipses is shown in Fig. 2b. In this representation it becomes evident that, although there is substantial overlap among Westerners and Easterners, there is a subtle trend in which Westerners and Paleoamericans occupy more positive values on the first PC, relative to the position of Easterners. Note also that Paleoamericans overlapped with Westerners more markedly than Easterners, reflecting a resemblance between some western groups and ancient remains from North America.
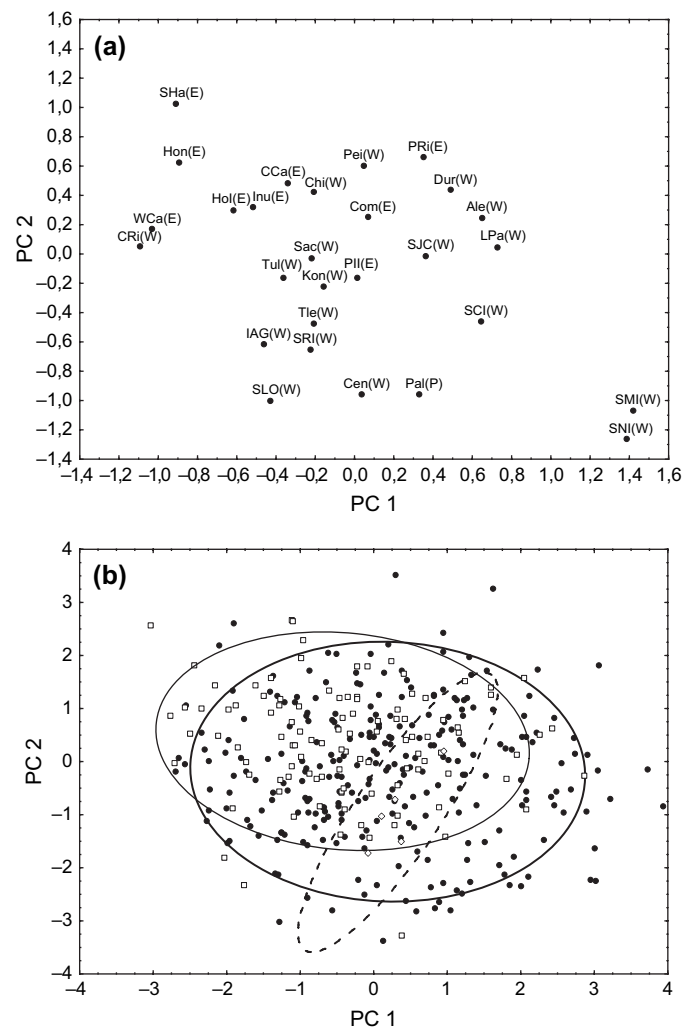

Fig. 2. Scatterplots based on the two first Principal Components. a) Group means. Westerner, Easterner, and Paleoamerican groups are represented by a W, E, and P, respectively. The points represent the mean score of each sample across PC 1 and 2. b) Distribution ellipses of 90% of variation for Westerners (thick line, solid circles), Easterners (thin line, open squares), and Paleoamericans (dotted line, open diamonds).

The Multidimensional Scaling graph (Fig. 3a), produced from the matrix of Mahalanobis distances among groups, provides a clear separation of western and eastern groups, and also shows the affinity of Paleoamericans with the western sub set of samples. Excepting the Isla Angel de la Guarda (western) sample, the more positive values across dimension 1 are occupied by Easterner groups. As expected, outlier groups as the Inuit and perhaps Puerto Rico and Honduras, are separated across dimension 2, not affecting the main East-West separation observed along dimension 1. Paleoamericans are now indisputably placed near the Westerner area of the graph. Note that when the matrix of Mahalanobis distances is computed after removal of the Paleoamerican sample (Fig. 3b), the East-West pattern remains clear and constant.

The Relethford and Blangero analysis, computed by pooling Westerners and Easterners (Table 3), demonstrated that Westerners have greater than average external gene flow, except in demographic scenarios that consider Westerner population

Table 3
Interregional Western (W) and Eastern (E) phenotypic distances[1]

		rii	obs	pred	res
E3:W1	E	0.006	0.962	0.985	−0.023
	W	0.091	0.969	0.900	0.069
E2:W1	E	0.012	0.962	0.982	−0.020
	W	0.066	0.969	0.929	0.041
E1:W1	E	0.029	0.962	0.967	−0.005
	W	0.032	0.969	0.964	0.005
E1:W2	E	0.050	0.962	0.941	0.020
	W	0.012	0.969	0.979	−0.010
E1:W3	E	0.063	0.962	0.925	0.036
	W	0.006	0.969	0.981	−0.012

[1] To the centroid ((rii), observed mean variance (obs), expected mean variance (pred), and residual variance (res), according to Relethford and Blangero (1990). Computations were made considering Eastern census size three and two times larger than the Western one (E3:W1 and E2:W1, respectively), the opposite scenario (E1:W3 and E1:W2), and equal census sizes (E1:W1). Heritability was set to $h^2 = 0.55$.

sizes two or three times larger than the Easterner populations. Further, the negative residuals in the Easterners suggest that migration into this region from an external source was more restricted than gene flow into the Western region, accepting equal population sizes or greater size in the East (the two most plausible options considering archaeological evidence). This pattern suggests that, in relation to the Easterners, the Westerners received a major number of migrants from some population other than those considered here.

Results concerning the computation of Fst without pooling of groups, following different demographic scenarios and considering a wide range of heritability values, are presented in Fig. 1.

The Fst's computed on all modern samples, assuming equal census sizes and an almost complete heritability (0.95), indicate that around 15% of the craniofacial variation was distributed among samples (Fst = 0.15, SE = 0.005). However, when the Fst is computed for only the Westerners, the values drop considerably (Fst = 0.11, SE = 0.006), while the Easterners gave higher values (Fst = 0.20, SE = 0.009). This pattern is maintained when different estimations of heritability are used, and regardless of the demographic scenario simulated, the total variation values remain consistent. If we assume that differences in heritability could have been operating among both regions, the only scenario in which Easterners are equal or less divergent than Westerners is when considering extremely low h^2 values in the West (0.25–0.35) and moderate to high h^2 values in the East (>0.45). Determining the extent of such differences in heritability values in natural human populations is beyond the scope of this paper. However, there is no *a priori* evidence supporting a strong disruption of the genetic basis of craniofacial variation during the settlement of the Americas, and hence, a scenario of greater Westerner differentiation. It seems more parsimonious to assume that differences in Easterner and Westerner heritability values are not so marked, even if they are not equal, and to look at the Fst's corresponding to intermediate heritability values (e.g., 0.35–0.65)
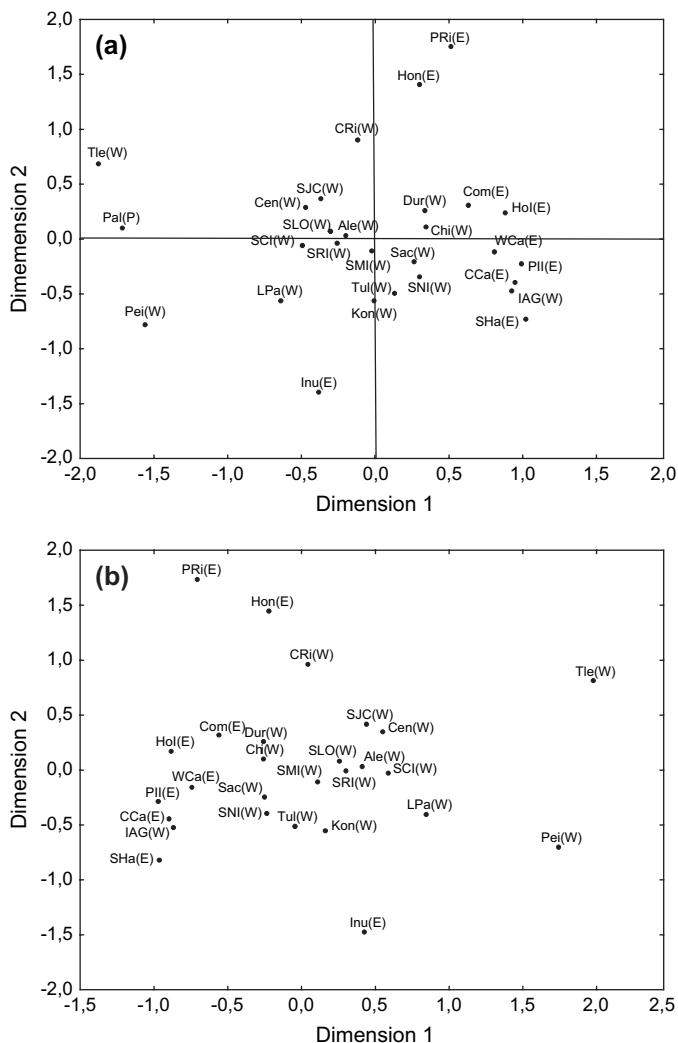

Fig. 3. a) Multidimensional scaling of the matrix of Mahalanobis distances among groups. Westerner, Easterner, and Paleoamerican groups are represented by a W, E, and P, respectively. Stress value: 0.143. b) The same analyses performed excluding the Paleoamericans. Stress value: 0.140.

rather than those with low heritability. If so, these figures suggest greater between-sample differentiation within the eastern sub set of samples in comparison to the more homogeneous Westerner pattern of inter group differences. Also, the pattern of intraregional differentiation remains the same even when extreme differences in demographic scenarios are used to compute the total Fst (Fig. 1). The standard errors calculated in the Relethford and Blangero method take sampling error into consideration, but they do not take into account evolutionary error (the variance of the among-population variance). If we assume that differentiation is mainly driven by stochastic processes like genetic drift, then multiple replicates of the same evolutionary history will result in a range of possible Fst. To have an estimate of the evolutionary variance for Fst in this particular sample, we calculated Fst for each principal component. The variance obtained was low (0.004, considering $h^2 = 0.55$) when compared to the range of Fst observed, thus suggesting that evolutionary error is quite negligible in the framework of Fst regional differences observed here.

Finally, as determined by Mantel tests, biological distances are correlated to both direct geographic distances among populations and geographic distances, considering a disruption among both population groups (Table 4). Necessarily, the GEOWE distances are correlated with the GEO distances ($r = 0,476$; p value < 0.00001). The correlation between biological distances and GEOWE could be due to their common dependence on geography (GEO). We therefore tested the partial correlation of Mahalanobis distances with GEOWE while keeping GEO constant (Table 4). When tested for significance by the Smouse-Long-Sokal test (Smouse et al., 1986), the partial correlation between BIO and GEOWE remain significant at $p = 0.011$. However, the correlation among BIO and GEO keeping GEOWE constant is not significant. Thus, these tests point towards a correlation among biological distances and geographic separation of both population groups above and beyond the predicted influences of direct distances among populations. In summary, the results of the Mantel tests suggest that a simple isolation by distance model is not enough to explain the craniofacial patterns observed.

Discussion

Our study adopts a double perspective: a comparison of modern and ancient samples, and a study of within and between-group variation and spatial patterning on modern samples, with the goal of estimating the likelihood of different microevolutionary processes in shaping phenotypic variability following the initial colonization event. Analyses performed to detect biological continuity or discontinuity among different periods are of crucial importance for understanding the biological manifestation of the numerous population shifts that certainly occurred in the early phases of the settlement of the New World. In addition, examination of within-group variation relative to inter group differences, as well as analyses of geographical patterning of genetic or phenotypic traits, can be of great help in understanding past historical and structural microevolutionary events. When dealing with extinct populations, craniofacial size and shape are usually analyzed in order to study diachronic changes and population structure and history in archaeological contexts. Although these markers are to some extent affected by non genetic factors, they present some advantages over other kinds of data when dealing with reconstruction of historical processes. For instance, when samples from different regions and periods are available, it is possible to apply different models specifically designed to test for the action of particular microevolutionary agents (Relethford and Blangero, 1990) or the existence of discontinuities across chronologic (Konigsberg, 1990; Relethford, 1991; Steadman, 2001; Stojanowski, 2004; Martínez-Abadías et al., 2005; Brace et al., 2006) or geographic (González-José et al., 2002, 2003; Brace et al., 2006) sequences.

In this context, this study evaluates craniofacial changes across time (using PCA comparisons of Paleoamericans and modern groups), and assesses modern patterns of differentiation (using Relethford and Blangero model and Fst statistics computed on modern groups) and geographical structure (using Mantel tests to compare biological and geographic distances). According to the Fst results, most of the craniofacial variation is distributed as differences among individuals within populations. In spite of this high level of intra population variability, a morphological trend could be observed in the pattern of inter-group distances that tends to separate Westerners and Paleoamericans from eastern groups (Figs. 2 and 3). Westerners and Paleoamericans display greater anterior and posterior neurocranial lengths, relative to Easterners. In addition to this pattern of inter group variability, the results suggest differences in terms the relative proportions of within-group and between-group diversity on either side of the continent. In fact, the Fst differences suggest different population dynamics in the two regions, with genetic drift the most parsimonious explanation for the high inter group differentiation levels in the East (high Fst). In contrast, gene flow may better explain the inter group homogenization and relative increase in within-group

Table 4
Mantel correlations[1]

Comparison	r	p
Correlations		
MAH vs. GEO	0.292	0.0172
MAH vs. GEOWE	0.318	0.0003
Partial correlations		
MAH vs. GEO.GEOWE	0.168	0.1152
MAH vs. GEOWE.GEO	0.213	0.0112

[1] p values obtained after 100,000 permutations. Among geographic distances (GEO, direct distances in kilometers); a matrix simulating low probability of contact among Westerner and Easterner population groups (GEOWE, direct distances in kilometers, measured passing by a waypoint in central Alaska for W-E elements); and Mahalanobis distances (MAH). Partial correlations according Smouse et al. (1986) are presented in the last two rows. MAH vs. GEO.GEOWE and MAH vs. GEOWE.GEO denotes comparison among MAH and GEO keeping the effects of GEOWE constant, and MAH and GEOWE keeping the effects of GEO constant, respectively. Differences among the jackknifed correlations (MAH vs. GEO and MAH vs. GEOWE) and partial correlations (MAH vs. GEO.GEOWE and MAH vs. GEOWE. GEO) were both significant ($t = 7.7$, $p < 0.00001$; $t = 18.4$; $p < 0.00001$, respectively).

variation in the West (low Fst). Furthermore, the Westerners as a whole appear to be more receptive to external gene flow than the Easterners (Table 3), at least considering the most parsimonious demographic scenarios (equal census sizes or greater effective population size in the East). When Fst's are computed taking into account the individual samples, Westerners always appear more variable than expected, regardless of the heritability values or demographic scenario considered (Fig. 1).

Rather than supporting a simple isolation-by-distance model influencing skull shape variation and acting in the continent as a whole, the Mantel correlation results suggest a hypothetical scenario in which craniofacial differences among any Westerner and Easterner group are proportional to their geographic distance via a waypoint in the northwest. This suggests a model of two different, more or less independent routes to the initial settlement of the Americas. In this context, the first subsidiary hypothesis cannot be rejected. The second subsidiary hypothesis, however, should be rejected, since in all the analyses that included Paleoamericans as well as Easterners

and Westerners, only the Westerners display some similarity with the Paleoamerican group (Figs. 2 and 3a).

To sum up, the craniofacial landscape of modern North Amerindians analyzed here would support the central hypothesis posed above, indicating an East-West differentiation, with the Westerners more similar to the most ancient remains, and the development of distinct population dynamics in each region (Fig. 4). Whether the East-West differentiation arose from historical, early events, like a stronger genetic presence of Paleoamericans along the Pacific corridor, or if the evolution of different microevolutionary scenarios on either side was sufficient to explain differentiation, remains unknown. However, some inferences pointing to the relative importance of each source of variation can be discussed, taking integrative data and previous research into account. Unfortunately, most previous studies concerning North American craniofacial variation have been centered on affinities among North Amerindians and ancient groups (Powell and Neves, 1999; Jantz and Owsley, 2001; González-José et al., 2003). In general
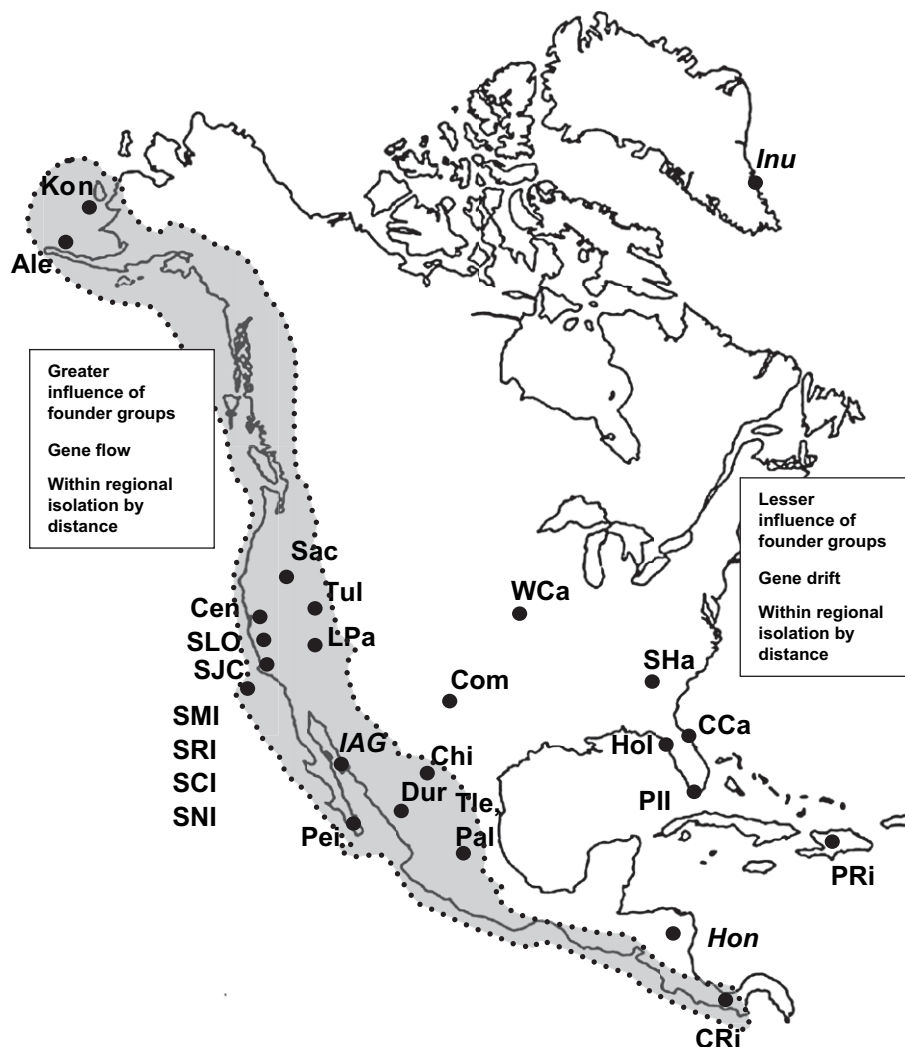

Fig. 4. Map of North America showing the populations studied. Shaded area depicts the putative extent of the Pacific coastal dispersal corridor, in terms of craniofacial affinities. Outlier groups in terms of craniofacial shape are presented in italics. Text boxes present the main putative historical and structural microevolutionary mechanisms suggested to have operated in each region.

terms, these studies reveal the high heterogeneity intrinsic to the ancient remains, as well as a lack of affinity between Paleoamericans and modern groups (e.g., Powell and Neves, 1999; Jantz and Owsley, 2001, with an exception in González-José et al., 2003). These studies are congruent with results presented in Fig. 3, where Paleoamericans are placed near to some particular modern groups.

Concerning the geographic patterning of craniofacial traits, we should note that the North-South gradient observed in dental traits by Greenberg et al. (1986) would not be expected to be as evident in the craniofacial spectrum of variation, being overwhelmed by the high levels of variation associated with the East-West separation. Also, East-West differences are not patterned in a smooth, gradient-like way, but in a way that probably reflects an early separation of both population groups. It is important also to note that studies of geographic structure are sensitive to differences in the arrangement of samples used, so making comparisons among our results and previous studies on this issue (Jantz et al., 1992; Nelson, 1998, 2006) is quite difficult. Finally, these results do not preclude some level of gene flow among Westerners and Easterners, which could have been important in some intermediate areas. However, a maintained difference in the amount of inter region relative to within-region gene flow is enough to generate the general trends depicted in our results.

Among the historical events that may have been responsible for the East-West differentiation, one of the most parsimonious ones is Dixon's (2001) hypothesis that the Pacific Coast acted as a narrow but ecologically continuous corridor, which would be inhabited first by Paleoamericans and later by their putative descendants, the Westerners. Dixon has proposed that the early occupation of the Americas obeyed a physiographic/ecological logic, and not a purely geographic logic. According to him, the major ecological zones of the Americas tend to be oriented linearly from North to South by vertical lines that parallel the major environmental zones, such as the Pacific Coast and the Western Cordillera. From this perspective, on a continental scale of analysis, the coast could be regarded as one of these zones because it constitutes an ecotone in which the resources of both marine and terrestrial biomes can be exploited. Consequently, gene flow in this ecological zone would be more intense than among adjacent zones because human displacement along the longitudinal ecological strips would be fast, even at similar latitudes. Supporting the scenario of an early Pacific dispersal route, some interesting resemblances among modern Pacific groups, like the Pericú, and the Paleoamerican series were recently reported by González-José et al. (2003).

In this context, Fix (2005) tested the likelihood of coastal versus terrestrial dispersal scenarios using simulation. He concluded that "Clovis hunters arriving through an ice-free corridor and expanding in a [North-South] 'blitzkrieg' wave was shown to be inconsistent with extensive genetic variability in Native Americans; a coastal migration route avoids this problem" (Fix, 2005:1). Dixon and Fix's coastal models could also explain why humans arrived at the tip of South America soon after they entered the New World in its extreme North, as the dates of Monte Verde now demonstrate (Dillehay, 1997; Dixon, 2001).

Interestingly, the patterns of cranial differentiation found here are in agreement with previous morphological studies carried out in South America. Neves (1989), for instance, compared Highland versus Lowland cranial samples by means of Principal Components Analysis applied to traditional craniometrics, and concluded that the two main clusters formed coincided with the geographical origins of the samples. In a recent analysis, Pucciarelli et al. (2006a) found that South American Westerners (including Pacific coastal populations) were well-differentiated from their counterparts to the East of the Andes. This dual pattern is also congruent with information derived from several genetic traits. Luiselli et al. (2000) suggested a clear East-West differentiation for the native South American populations, based on 16 genetic systems. According to Tarazona-Santos et al. (2001), the Andean peoples showed higher levels of intra population variation and lower levels of inter population variation when compared to lowlanders. Thus, they proposed an evolutionary model for South Amerindian males resting on differential patterns of genetic drift and gene flow on each side of the Cordillera. On the West side, populations had a larger effective size and higher gene flow levels than in the East, favoring a trend towards inter group homogenization of the genetic pool. For Fuselli et al. (2003), eastern populations settled in Amazonia and Chaco regions exhibit high rates of genetic drift and low rates of gene flow, evoking a trend towards inter group genetic differentiation. Although they were unable to identify a common genetic pattern for South America as a whole, when both side populations were analyzed separately, the mitochondrial diversity fit well in the isolation by distance model, suggesting independent evolutionary dynamics for each side.

The dual pattern of differentiation found in South America was, although indirectly, also supported by archaeological information. It is well known that social and technological developments differed sharply on each side of the Cordillera, Westerners being more advanced in technology, food production, social organization, and population density than Easterners (Meggers, 1979; Bruhns, 1994). Mandatory population displacement and long distance exchange of goods, for example, could have been enhanced by complex social and economic organizations, explaining the higher rate of gene flow among highlanders. A similar explanation, however, seems inadequate to explain the same dual pattern in Central and North America, since in the northern continent, such high levels of social development and cultural complexity were not reached.

Our results suggest that, despite the differences of cultural development, the same biological differentiation process may have occurred in both continents. The northern mountain chains, such as the Rocky Mountains in the USA, and Sierra Madre in Mexico, are not part of one continuous barrier, but perhaps they were enough to preclude a free East-West transportation. Perhaps, an interesting alternative is not to focus on geographic barriers, but on the existence of ecological corridors, as a way to preclude gene flow among groups inhabiting different ecological niches.

Since all of the East and West samples analyzed here represent pre-contact prehistoric populations, we propose that

the ecological logic for human settlement and dispersal suggested by Dixon (2001) for the early Americans also prevailed in the continent until very late in time. Moreover, the results presented here may point to different degrees of contribution of Paleoamerican genes—greater in the West and lower in the East—fuelled by the intensive use of a Pacific corridor, which could result in the dual pattern found in South, Central, and North America (e.g., a pan-American phenomenon). Additionally, differences in the relative amount of genetic drift and gene flow in both regions might also act to increase, or at least conserve, the East-West differences. Accordingly, these elaborations cannot be based on geographical, ecological, and socio-cultural characteristics unique to South America (Pucciarelli et al., 2006a), but should rely otherwise on characteristics of continental scale.

Finally, although some level of response to selective pressures is probably involved in the skull differences we detected here, recent studies have demonstrated that adaptive explanations alone cannot be invoked to account for modern human craniofacial variation (Relethford, 2002; Roseman, 2004; Roseman and Weaver, 2004; see Culotta, 2005 for a discussion). Considering that the settlement of the Americas was of continental scale and involved at least 12,000 years, realistic explanations for the differences observed may involve alternative microevolutionary agents other than exclusively selection, such as the differential rate of genetic drift/gene flow on either side of the continent, as shown in the present study.

Conclusions

An East-West differentiation seems to be the main pattern of morphological separation among Central and North American Indians. A similar pattern has been shown in South America (Pucciarelli et al., 2006a). As argued here, this pattern may result from historic processes such as the ancient and intensive use of a Pacific dispersal corridor, as well as the effects of differential gene drift/gene flow, with genetic drift having had a greater influence in the East, and gene flow a greater influence in the West. In addition, the observed craniofacial patterning supports a settlement scenario of two different, and to some extent isolated, routes of initial dispersal across the Americas. Further studies relying on larger numbers of New World cranial samples will be of paramount importance for testing whether the other ecological longitudinal strips suggested by Dixon (2001) were as important as the Pacific Coast and the Cordillera in structuring the human biological diversity in the Americas.

Acknowledgements

Appendix. Supplementary material

Supplementary data associated with this article can be found, in the online version, at 10.1016/j.jhevol.2007.08.011.

References

Bass, W.M., 1964. The variation in physical types of the prehistoric Plains Indians. Plains Anthropol. 9, 65–145. Memoir (No 1).

Brace, C.L., Hunt, K.D., 1990. A non-racial craniofacial perspective on human variation: A(ustralia) to Z(uni). Am. J. Phys. Anthropol. 83, 341–460.

Brace, C.L., Nelson, A.R., Seguchi, N., Oe, H., Sering, L., Quifeng, P., Yongy, L., Tumen, D., 2001. Old World sources of the first New World human inhabitants: a comparative craniofacial view. Proc. Natl. Acad. Sci. 98 (17), 10017–10022.

Brace, C., Nelson, A.R., Pan, Q., 2004. Peopling of the New World: a comparative craniofacial view. In: Barton, C., Clark, G., Yesner, D., Pearson, G. (Eds.), The Settlement of the American Continents: A Multidisciplinary Approach to Human Biogeography. Arizona University Press, Tucson, pp. 28–38.

Brace, C.L., Seguchi, N., Quintyn, C.B., Fox, S.C., Nelson, A.R., Manolis, S.K., Qifeng, P., 2006. The questionable contribution of the Neolithic and the Bronze Age to European craniofacial form. Proc. Natl. Acad. Sci. U.S.A. 103, 242–247.

Bruhns, K.O., 1994. Ancient South America. Cambridge University Press, Cambridge.

Carneiro, R.L., 1970. A theory of the origin of the state. Science 169, 733–738.

Culotta, E., 2005. What genetic changes made us uniquely human? Science 309, 91.

Dillehay, T.D., 1997. Monte Verde. A Late Pleistocene Settlement in Chile. Vol. 2: The Archaeological Context and Interpretation. Smithsonian Institution Press, Washington.

Dixon, E.J., 2001. Human colonization of the Americas: timing, technology and process. Quatern. Sci. Rev. 20, 277–299.

Dressino, V., Pucciarelli, H.M., 1999. Growth of functional cranial components in *Saimiri sciureus boliviensis* (Cebidae): a longitudinal study. Growth, Development and Aging 63, 111–127.

Fix, A.G., 2005. Rapid deployment of the five founding Amerind mtDNA haplogroups via coastal and riverine colonization. Am. J. Phys. Anthropol. 128, 430–436.

Focacci, G., Erices, S., 1973. Excavaciones en túmulos de San Miguel de Azapa. Actas IV Congreso de Arqueología Chilena, Número Especial, pp. 44–55.

Fuselli, S., Tarazona-Santos, E., Dupanloup, I., Soto, A., Luiselli, D., Pettener, D., 2003. Mitochondrial DNA diversity in South America and the genetic history of Andean highlanders. Mol. Biol. Evol. 20, 1682–1691.

González-José, R., García-Moro, C., Dahinten, S., Hernández, M., 2002. The origin of the Fueguian Patagonians: an approach to population history and population structure using R matrix and matrix permutation methods. Am. J. Hum. Biol. 14, 308–320.

González-José, R., González-Martín, A., Hernández, M., Pucciarelli, H.M., Sardi, M., Rosales, A., van der Molen, S., 2003. Craniometric evidence for Palaeoamerican survival in Baja California. Nature 425, 62–65.

González-José, R., Neves, W., Lahr, M., González, S., Pucciarelli, H., Hernández Martínez, M., Correal, G., 2005. Late Pleistocene/Holocene craniofacial morphology in Mesoamerican Paleoindians: implications for the people of the New World. Am. J. Phys. Anthropol. 128, 772–780.

Greenberg, J.H., Turner, C.G., Zegura, S.L., 1986. The settlement of the Americas: a comparison of the linguistic, dental, and genetic evidence. Curr. Anthropol. 27, 477–497.

Harpending, H.C., Ward, R.H., 1982. Chemical Systematics and Human Populations. In: Nitecki, M. (Ed.), Biochemical Aspects of Evolutionary Biology: Genetic Structure of Small Populations. University of Chicago Press, Chicago, pp. 213–261.

Howells, W.W., 1973. Cranial Variation in Man: a Study by Multivariate Analysis of Patterns of Difference among Recent Human Populations. Papers of the Peabody Museum of Archaeology and Ethnology. Harvard University Press, Cambridge.

Howells, W.W., 1989. Skull Shapes and the Map: Craniometric Analysis in the Dispersion of Modern Homo. Papers of the Peabody Museum Archaeology and Ethnology. Harvard University Press, Cambridge.

Jantz, R.L., Hunt, D.R., Falsetti, A.B., Key, P.J., 1992. Variation among North Amerindians: Boas anthropometric data. Hum. Biol. 64 (3), 271–279.

Jantz, R.L., Owsley, D.W., 2001. Variation among early North American crania. Am. J. Phys. Anthropol. 114, 146–155.

Klaauw, C.J. van der, 1948–52. Size and position of the functional components of the skull. Archaeol. Neerl. Zool. 9, 1–559.

Konigsberg, L.W., 1990. Analysis of prehistoric biological variation under a model of isolation by geographic and temporal distance. Hum. Biol. 62, 49–70.

Krauss, M.E., 1980. Alaska native languages: past, present and future. Alaska Native Language Center Research, Paper 4, Fairbanks.

Lilliard, J.B., Heizer, R.F., Fenenga, F., 1939. An Introduction to the Archaeology of Central California. Sacramento Junior College, Department of Anthropology, Sacramento. Bulletin 2.

Llop, E., Harb, Z., Moreno, R., Rothhammer, R., 2002. Genetic marker variation in coastal populations of Chile. Homo 53, 170–177.

Luiselli, D., Simoni, L., Tarazona-Santos, E., Pastor, S., Pettener, D., 2000. Genetic structure of Quechua-speakers of the central Andes and geographic patterns of gene frequencies in South Amerindian populations. Am. J. Phys. Anthropol. 113, 5–17.

Manly, B.F.J., 1994. Multivariate Statistical Methods. Chapman and Hall, Londres.

Mantel, N., 1967. The detection of disease clustering and a generalized regression approach. Cancer Res. 27, 209–220.

Marroig, G., Cheverud, J., 2005. Size as a line of least evolutionary resistance: diet and adaptive morphological radiation in New World Monkeys. Evolution 59, 1128–1142.

Martínez-Abadías, N., González-José, R., González-Martín, A., van der Molen, S., Talavera, A., Hernández, P., Hernández, M., 2005. Phenotypic evolution of human craniofacial morphology after admixture: a geometric morphometrics approach. Am. J. Phys. Anthropol. 129, 387–398.

Meggers, B.J., 1979. Prehistoric America: An Ecological Perspective. De Gruyter, Aldine.

Moss, M.L., 1961. Rotation of the otic capsule in bipedal rats. Am. J. Phys. Anthropol. 19, 301–307.

Moss, M.L., 1997a. The functional matrix hypothesis revisited. 2. The role of an osseous connected cellular network. Am. J. Orthod. Dentofac. Orthop. 112, 221–226.

Moss, M.L., 1997b. The functional matrix hypothesis revisited. 3. The genomic thesis. Am. J. Orthod. Dentofac. Orthop. 112, 338–342.

Moss, M.L., 1997c. The functional matrix hypothesis revisited. 4. The epigenetic antithesis and the resolving synthesis. Am. J. Orthod. Dentofac. Orthop. 112, 410–417.

Moss, M.L., Young, R.W., 1960. A functional approach to craniology. Am. J. Phys. Anthropol. 18, 281–292.

Muñoz, Y., Focacci, G., 1983. Asentamientos aldeanos en los valles costeros de Arica. Universidad de Tarapacá. Documentos de Trabajo 3, 1–43.

Nelson, A.R., 1998. A Craniofacial Perspective on North American Indian Population Affinities and Relations. Anthropology. Michigan University Press, Ann Arbor.

Nelson, A.R., 2006. Patterns of Craniometric Variation and Geographical Distribution in North America: a Historical Comparison. In: Bonnichsen, R., Lepper, B.T., Stanford, D., Watters, M.R. (Eds.), Paleoamerican Origins: Beyond Clovis. Center for the Study of the First Americans. Texas A&M University Press, Texas, pp. 205–216.

Neumann, G.K., 1942. The origin of the Prairid physical type of American Indian. Papers Michigan Acad. Sci. Arts Lett. 27, 539–542.

Neumann, G.K., 1952. Archaeology and Race in the American Indian. In: Griffin, J.B. (Ed.), Archaeology of Eastern United States. Chicago University Press, Chicago, pp. 13–34.

Neves, W.A., 1989. A ocupação pré-colonial da América do Sul. Reflexões a partir de um exercício bio-antropológico. Boletim do Museu Paraense Emílio Goeldi. Série Antropologia 5, 79–103.

North, A.W., 1908. The native tribes of Lower California. Am. Anthropol. 10, 236–250.

Powell, J.F., Neves, A.W., 1999. Craniofacial morphology of the First Americans: pattern and process in the peopling of the New World. Yearb. Phys. Anthropol. 42, 153–188.

Pucciarelli, H.M., Dressino, V., Niveiro, M.H., 1990. Changes in skull components of the squirrel monkey evoked by growth and nutrition: an experimental study. Am. J. Phys. Anthropol. 81, 535–543.

Pucciarelli, H.M., Neves, W.A., González-José, R., Sardi, M.L., Ramírez Rozzi, F., Struck, A., Bonilla, M.Y., 2006a. East-West cranial differentiation in pre-Columbian human populations of South America. Homo 57, 133–150.

Pucciarelli, H.M., Ramírez Rozzi, F.V., Muñe, M.C., Sardi, M.L., 2006b. Variation of functional cranial components in six Anthropoidea species. Zoology 109, 231–243.

Pucciarelli, H.M., Sardi, M.L., Jimenez López, J.C., Serrano, C., 2003. Early peopling and evolutionary diversification in America. Quatern. Int. 109–110, 123–132.

Relethford, J.H., 1991. Genetic drift and anthropometric variation in Ireland. Hum. Biol. 63, 155–165.

Relethford, J.H., 1996. Genetic drift can obscure population history: problem and solution. Hum. Biol. 68, 29–44.

Relethford, J.H., 2002. Apportionment of global human genetic diversity based on craniometrics and skin color. Am. J. Phys. Anthropol. 118, 393–398.

Relethford, J.H., Blangero, J., 1990. Detection of differential gene flow from patterns of quantitative variation. Hum. Biol. 62, 5–25.

Relethford, J.H., Crawford, M.H., Blangero, J., 1997. Genetic drift and gene-flow in post famine Ireland. Hum. Biol. 69, 443–465.

Relethford, J.H., Harpending, H.C., 1994. Craniometric variation, genetic theory and modern human origins. Am. J. Phys. Anthropol. 95, 249–270.

Roseman, C.C., 2004. Detecting interregionallity diversifying natural selection on modern human cranial form by using matched molecular and morphometric data. Proc. Natl. Acad. Sci. 101, 12824–12829.

Roseman, C.C., Weaver, T.D., 2004. Multivariate apportionment of global human craniometric diversity. Am. J. Phys. Anthropol. 125, 257–263.

Sardi, M.L., Ramírez Rozzi, F., Pucciarelli, H.M., 2004. The Neolithic transition in Europe and North Africa: the functional craniology contribution. Anthropol. Anz. 62, 129–145.

Sassman, K.E., Anderson, D.G., 2004. Late Holocene period, 3750-650 B.C. The little Sycamore site in the early Milling Stone cultures of Southern California. Am. Antiq. 20, 112–123.

Schluter, D., 1996. Adaptive radiation along genetic lines of least resistance. Evolution 50, 1766–1774.

Simoni, L., Tarazona-Santos, E., Luiselli, D., Pettener, D., 2000. Genetic Differentiation of South American Native Populations Inferred from Classical Markers: from Explorative Analysis to a Working Hypothesis. In: Renfrew, C. (Ed.), America Past, America Present: Genes and Languages in the Americas and Beyond. McDonald Institute for Archeological Research, Cambridge, pp. 123–134.

Smouse, P.E., Long, J.C., Sokal, R.R., 1986. Multiple regression and correlation extensions of the Mantel test matrix correspondence. Syst. Zool. 35, 627—632.

Steadman, D.W., 2001. Mississippians in motion? A population genetic analysis of interregional gene flow in West-Central Illinois. Am. J. Phys. Anthropol. 114, 61—73.

Stojanowski, C.M., 2004. Population history of native groups in pre- and post-contact Spanish Florida: aggregation, gene flow, and genetic drift on the South-Eastern U.S. Atlantic coast. Am. J. Phys. Anthropol. 123, 316—332.

Tarazona-Santos, E., Carvalho-Silva, D.R., Pettener, D., Luiselli, D., De Stefano, G.F., Martinez Labarga, C., Rickards, O., Tyler-Smith, C., Pena, S.D.J., Santos, F.R., 2001. Genetic differentiation in South Amerindians is related to environmental and cultural diversity: evidence from the Y chromosome. Am. J. Hum. Genet. 68, 1485—1496.

Wedel, W.R., 1941. Archaeological Investigations at Buena Vista Lake, Kern County, California. Bureau of American Ethnology, Washington. Bulletin 130.

Wilkinson, L., Engelman, L., Corter, J., Coward, M., 2002. Statistics I. Systat Software Inc., Richmond.