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Carla Santana
Laboratorio Biologia Molecular Diagnóstica (BIMODI), Querétaro, Qro, México

Gino Noris
Laboratorio Biologia Molecular Diagnóstica (BIMODI), Querétaro, Qro, México

Marco Antonio Meraz-Ríos
Departamento de Biomedicina Molecular, Cinvestav-IPN, Mexico

Jonathan J. Magaña
Laboratorio de Medicina Genómica, Departamento de Genética, Instituto Nacional de Rehabilitación, Secretaria Salud, Mexico

Emma S. Calderon-Aranda
Departamento de Toxicologia, Cinvestav-IPN, Mexico

See next page for additional authors

Authors
Carla Santana, Gino Noris, Marco Antonio Meraz-Ríos, Jonathan J. Magaña, Emma S. Calderon-Aranda, Leonor C. Acosta-Saavedra, Maria de Lourdes Muñoz, and Rocio Gómez

Genetic analysis of 17 Y-STRs in a Mestizo population from the Central Valley of Mexico

Carla Santana[1], Gino Noris[1], Marco Antonio Meraz-Ríos[2], Jonathan J. Magaña[3], Emma S. Calderon-Aranda[4], Leonor C. Acosta-Saavedra[4], Maria de Lourdes Muñoz[5], Rocio Gómez[4]

[1]Laboratorio Biologia Molecular Diagnóstica (BIMODI), Querétaro, Qro, México

[2]Departamento de Biomedicina Molecular, Cinvestav-IPN, Mexico

[3]Laboratorio de Medicina Genómica, Departamento de Genética, Instituto Nacional de Rehabilitación, Secretaria Salud, Mexico

[4]Departamento de Toxicologia, Cinvestav-IPN, Mexico

Correspondence to: Rocío Gómez, Ph.D., Departamento de Toxicología, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional. Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco, 07360, México, D.F., México. E-mail: mrgomez@cinvestav.mx.

Key words: Y-STRs, Mexican Mestizo population, Forensic, Amerindian lineages, Population genetics

Abstract

This study aims to portray the complex diversity of the Mexican Mestizo population, which represents 98.8% of the entire population of Mexico. We compiled extended haplotype data of the Y chromosome from populations in the Central Valley of Mexico (CVM), which were compared to other Mestizo and parental (Amerindian, European and African) populations. A complex ancestral relationship was found in the CVM population, suggesting cosmopolitan origins. Nevertheless, the most preeminent lineages point towards a European ancestry, where the R1b was the most frequent. In addition, important frequencies of Amerindian linages were also found in the Mestizo sample studied. Interestingly, the Amerindian ancestry showed a remarkable substructure, which was represented by the two main founding lineages: QL54 (x M3) and M3. However, even within each lineage a high diversity was found despite the small number of samples bearers of these lineages. Further, we detected important genetic differences between the CVM populations and the Mexican Mestizo populations from the north and south. This result points to the fact that Mestizo populations present different ancestral proportions, which are related to the demographic events that gave origin to each population. Finally, we provide additional forensic statistical parameters that are useful in the interpretation of genetic analysis where autosomal loci are limited. Our findings illustrate the complex genetic background

of the Mexican Mestizo population and reinforce the need to encompass more geographic regions to generate more robust data for forensic applications.

Mexican Mestizos are a complex population that emerged 520 years ago with the miscegenation of Amerindian populations by Europeans (Iberian Peninsula) (Beezley 2011). Also, a continuous gene flow of African slaves was conducted from the 16^{th} to the 18^{th} centuries (Basu et al. 2008). Meanwhile, multiple migratory waves contributed with a constant influx of men from Europe and Africa, increasing the patrilineal genetic heterogeneity. In addition to transatlantic migrations, demographic events such as bottleneck, founder effect, local drift and rapid population growth create differences in Y chromosome haplotype frequencies along the country. Consequently, heterogeneous genetic patterns were produced. In general, this heterogeneity shows a south-north clinal increase of European ancestry (Luna-Vazquez et al. 2008). On the other hand, the Amerindian lineage shows a southward distribution, whereas the African ancestry exhibits low frequencies and heterogeneous distribution patterns (Martinez-Cortes et al. 2012; Salazar-Flores et al. 2010). This genetic variability justifies the genetic characterization of diverse geographical Mexican populations.

Short tandem repeat (STR) polymorphisms in the non-recombining region (NRY) of the Y chromosome (Y-STR) are passed down patrilineal generations unchanged, except for random mutations events (Buttler 2011; National research Council 1996). The fact that these markers that are present only in males make them powerful tools in forensic DNA testing, especially in sexual assault, in which autosomal STRs show high levels of female DNA (Butler 2011). However, since Y chromosome genetic profiles are shared by paternal relatives, population forensic parameters like Y-STR profiles frequency and discrimination power, must be determined to avoid statistical errors (National research Council 1996). These shearings could have more implications in inbreed or young populations such as the Mexican Mestizo, which emerged only nine to fifteen generations ago (Johnson et al. 2011).

Along with the forensic implications, uniparental markers are considered the best genetic system to trace the history of human migrations (Salzano 2007). Although several genetic Y-STRs databases are currently available, these databases contain information restricted to minimal haplotype loci (Luna-Vazquez et al. 2008; Padilla-Gutierrez et al. 2008; Rangel-Villalobos et al. 2001; Salazar-Flores et al. 2010). Therefore the use of a bigger set of Y-STRs is desirable as well as the use of chromosome Y-SNPs markers to obtain greater precision, which can also be used to compare different populations to establish their relationships. In addition, regional differences, admixture, and demographic events require the examination of different samples from the same population to obtain a comprehensive representation of the genetic complexity, which is a fundamental issue in genetic anthropology and forensic sciences (Salazar-Flores et al. 2010; Martinez-Cortes et al. 2012).

Thus, the main aims of this article were: (1) to study the genetic composition of the Mestizo population in the Central Valley of Mexico (CVM) and to compare this population with others, and (2) to estimate the statistical forensic parameters using 17 Y-STRs loci routinely employed in anthropological, forensic and population genetics. Our findings suggest that the Mestizo population from the Central Valley area is a cosmopolitan population with specific genetic characteristics. Among these characteristics, we found high frequency of the linage R1b, which is related to Western Europe. Further, the second ancestry found was the Q lineage, which showed a striking genetic substructure represented by the sub-lineages QL54 and QM3. Our data highlights the importance of determining local-specific patterns throughout the country to establish the complex genetic background of the Mexican Mestizo population. In addition, this data could support the forensic parameters that will enable the clarification of kin relations.

Materials and Methods

Population of study

Blood samples were collected from 231 unrelated men belonging to the Mexican Mestizo population having at least three generations of ancestors born in Mexico. The studied population was recruited from Mestizos living in the Central Valley of the country (North-Central and East-Central regions) (Figure 1). This population included 121 men from Querétaro and 63 men from Guanajuato (North-Central region), as well as 47 men from Puebla (East-Central region).

Each individual signed an informed consent validated by the Ethics Committee of the Bimodi's Research Unit. In addition, genealogical data were also obtained from each person to ensure that the individuals were unrelated through at least three generations.

Molecular Analysis

Y-chromosome haplotyping

Genomic DNA was extracted from peripheral blood leukocytes using Qiamp DNA Mini Kit (Qiagen, Düsseldorf, Germany). The non-recombining region of the Y chromosome (NRY) was characterized for each man by seventeen Y-chromosome short tandem repeats markers (DYS19, DYS385a/b, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS437, DYS438, DYS439, DYS448, DYS456, DYS458, DYS635, and GATA-H4) using the commercial typing AmpF*l*STR Yfiler™ PCR amplification kit (Applied Biosystems, Carlsbad, CA, USA), according to the manufacturer's instructions, and a Veriti 96-Well Fast Thermal Cycler (Applied Biosystems, Carlsbad, CA, USA). The resulting amplicons were analyzed by electrophoresis on the ABI Prism 3130XL Genetic Analyzer using the GeneMapper ID v.3.2. software (Applied Biosystems, Carlsbad, CA, USA).

Statistical and phylogenetic analysis

Population genetics parameters

Allele and haplotype frequencies, number of alleles (*k*), haplotype diversity (HD), genetic diversity over loci (*h*), and mean pairwise differences were estimated using Arlequin v. 3.5 software (Excoffie et al. 2010). Number of unique haplotypes (nuh) was estimated by direct counting. The following forensic parameters were determined: discrimination capacity (DC) was calculated by the expression: DC=h/n, where "h" is the total number of different haplotypes and "n" is the total number of individuals in the sample; the power of discrimination (pD) was considered equivalent to genetic diversity, and matching probability (MP) was obtained from the equation: MP=1-HD.

Y-chromosome haplogroups

A haplogroup predictor was used to assign Y-STR haplotypes into Y-chromosome haplogroups (http://www.hprg.com/hapest5/) (Athey 2006). In order to confirm and characterize the Amerindian haplogroups found through the software predictor, five single nucleotide polymorphic (SNPs) markers were genotyped. These polymorphisms included the following SNPs: M242, MEH2, M346, L54, and M3. All SNPs were genotyped with C1000 Thermal cycler (Bio-Rad Life Science, Hercules, CA, USA) using TaqMan assays (Applied Biosystems, Carlsbad, CA, USA). The high resolution haplogroups were assigned according to the SNP haplotyping following the most update Y-chromosome nomenclature (Karafet et al. 2008). The combination of SNP and STR markers determined the paternal lineages in the studied individuals.

Network analysis

The phylogenetic relationship, the diversity patterns as well as the ages of haplogroups through coalescence time estimation were constructed using a median-joining (MJ) method with Network v. 4.6.1.2 software (Bandelt et al. 1999), and with Network Publisher software. For coalescence time estimates, a rate of one mutation every 453 years was used, which was estimated by taking the inverse per generation mutation rate of each locus multiplied by the number of loci and by generation time, or 25 years (Chandler 2006). The NRY haplotypes used to generate the networks consisted of 15-YSTRs. DYS385ab locus was excluded from the network analysis because it represents a duplicate STR locus, and these loci were also excluded from all the analysis performed. The Y-STRs loci were weighted based on the data reported previously (Roewer et al. 2013).

Comparison with other populations

In order to compare our data with other populations, haplotype information was collected from previous reports in Mexican Mestizo populations. A total of forty-four populations (n=2498) were included in the database and used for further analysis (Supplementary Information, Table S1). Genetic relationships of the studied populations and between other Mestizo populations, as well as with other parental populations (Amerindian, European and African) were analyzed based on genetic distances (R_{ST} and F_{ST} values using 5000 permutations) with the Arlequin v.3.5 software (Excoffie et al. 2010), and visualized by the multidimensional scaling plot (MDS) with the SPSS v.11 program.

Population subdivision was assessed by Analysis of Molecular Variance (AMOVA) using geography as subdivision criterion with Arlequin Software v.3.5 (Excoffie et al. 2010). Genetic relationships in our populations as well as among continental populations were analysed base on the genetic distances (R_{ST} and F_{ST}, using 1000 permutations) with Arlequin Software v.3.5 (Excoffie et al. 2010), and visualized by multidimensional scaling plot (MDS) with SPSS v.11.

Quality control

Control DNA 007 was used as international validated internal control (Applied Biosystems, Carlsbad, CA, USA).

Results

Haplotype diversity

The haplotypes of the 17 Y-STRs loci in the 231 individuals studied are shown in the Supplementary Information (Table S2). We observed 230 different haplotypes using 17 Y-STRs markers (from 231 samples), which meant that almost every sample is unique and only two individuals (Guanajuato and Querétaro) share the same haplotype. As a consequence, high haplotype diversity was found in the three studied populations (Guanajuato, GTO; Puebla, PUE; and Querétaro, QRO) as well as in the population as a whole. Regarding other combinations using <10 Y-STRs, we found that the most frequent haplotype combination was 14-11-14-13-29-24-11-13 (DYS19-DYS385a-DYS385b-DYS389I-DYS389II-DYS390-DYS391-DYS392), which represented almost 5% of all haplotypes. Other combinations are shown in the Supplementary Information (Table S3).

The distribution of allele frequencies of the 17 loci and the number of different alleles (*k*) are shown in Table 1. With regard to the allelic combination in the DYS385a/b locus, we found that the more frequent genotypes were 11-14 (GTO=0.286, PUE=0.149, QRO=0.158) and 15-17 (PUE=0.128) (Supplementary Information, Table S4). Finally, the locus diversity over loci as well as mean number of pairwise differences, were similar across populations coming from different areas (Supplementary Information, Table S5).

AMOVA and pairwise differentiation tests

In order to assess the heterogeneity among populations in the three states, an AMOVA test was performed. The results showed a non-significant heterogeneity among populations (*P*=0.551). In contrast, the highest variation was found within populations (~ 99%). In addition, a pairwise population differentiation test was performed which yielded non-significant differences among populations ($0.220 \leq P \leq 0.750$) confirming the findings obtained with the AMOVA. Since non-significant genetic differences were found among the three populations, henceforth, this population will be referred to as a whole and will be identified as CVM (Central Valley of Mexico).

Y-chromosome haplogroups

In order to know the main patrilineal lineages that are present in the Mexican Mestizo population, haplogroups were assigned and their frequency was calculated, using Bayesian probability with Haplogroup predictor software. Our results showed that the most frequent haplogroups were R1b (~ 0.41) and Q (~ 0.21). In addition, lineages such as E1b1b (0.07) and J1 (0.06) were also found. This data suggests an important genetic substructure in the Mestizo population, which is principally represented by the Amerindians (Q) and Europeans (R1b) lineages. It is worth mentioning that an important diversity was evident in the most frequent haplogroups (Supplementary Information, Table S2).

In order to elucidate the diversity within the Amerindian lineage a more accurately analysis was done. The bearers of the Q lineage were confirmed using the single nucleotide polymorphisms: M242, MEH2, M346, QL54 and QM3. As expected, the most frequent sub-lineage was QM3, which was found with a frequency of 77%, whereas QL54 was found in ~ 23% of the Mestizo haplotypes.

Network analysis

To establish the diversity within the Q haplogroup, a median joining network was constructed for each sub-haplogroup (QL54 and QM3) using 15 Y-STRs (Supplementary Information, Table S1). Both sub-lineages showed a star-like network where different clusters were identified. These highly diverse patterns, suggest that both sub-lineages are relatively young or that they could have suffered different demographic events (e.g. bottleneck, extensive isolation, genetic drift, and founder effect). In addition, it is worth nothing the length of the branches, where up to ten mutations were found. About the lineage QL54, of note is the high diversity within the populations from QRO and PUE, which show high variability in their haplotypes, despite the small number of samples. The QM3 lineage shows high variability in the haplotypes of each geographical region, suggesting even higher diversity within this lineage (Figure 2).

Comparison with other populations

In order to find the ancestral relationship between the sub-lineages found in the Mestizo population, a comparison with previously reported data was done. This analysis included Asian and Amerindian populations from North America related to the First Nations of Canada and Alaska (Gwich'in and Mi'kmaq), Western Canadian Inuit (Inuvialuit), Canadian Metis, Federal Recognized Tribes (Wiyot) and Pacific Northwest Coast of North America (Tlingit). All analyses were carried out using 15 Y-STRs (Supplementary Information, Table S1).

Regarding QL54, this lineage is connected with the ancestral populations from the Tuva Republic and Northeast Siberia through 16 mutations. Moreover, this lineage presents at least two clusters. The first cluster shows a genetic relationship between the Mestizo populations from QRO and PUE and the Gwich'in and Mi'kmaq ethnic groups. The second cluster displayed the haplotypes from the CVM population (Supplementary Information, Figure S1).

With regard to the QM3 lineage, it shows an ancestral relationship to Asian populations from Northeast Siberia, which are separated by at least 11 mutations of Mexican Amerindian lineages. QM3 presents a high diversity, wherein are distinguished at least three clades. The first clade is represented by the populations from North America (Gwich'in, Inuvialuit, Tlingit,) and Northeast Siberia, which show an ancestral relationship with a few haplotypes from the CVM population. The second clade shows the ancestral relation between Mexican Mestizos and North American populations such as Canadian Metis, Tlingit and Wiyot. Finally, the third clade is represented by the diversity of the Mexican Mestizo population (Supplementary Information, Figure S2).

On the other hand, an AMOVA test was performed in order to compare CVM data with Southern (Yucatan and Chiapas), Western (Jalisco) and North-Central (Guanajuato) regions of Mexico using 17 Y-STRs reported by previous studies (Salazar-Flores et al. 2010). The AMOVA test showed important variations among populations ($P \leq 0.001$). However, this variation markedly diminished when Southern populations were excluded ($P \geq 0.050$). The pairwise population differentiation test showed statistical differences between CVM, Western and North Central populations as well as Southern populations ($P \leq 0.001$), corroborating the genetic structure patterns detected with the AMOVA test.

Moreover, an MDS plot was performed with haplotype R_{ST}, which showed a clear separation between Southern populations and the rest (Figure 3). In addition, we also compared it with other populations such as Aguascalientes (Center-North) and Mexico City, using the minimal haplotype (DYS19, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393 and DYS385a/b) (Supplemental Information, Figure S3).

To ascertain whether this substructure is related to Amerindian ancestry, we compared the data with other Amerindian populations, which present the most preeminent founding lineages. These populations include Amerindian populations from Mexico, Guatemala and North America; all analyses were carried out using 15 Y-STRs (Supplemental Information, Table S1). The AMOVA test showed a scanty difference between Southern (Chiapas and Yucatan) and Amerindian populations ($F_{ST} \sim 0.0068$, 0.68%), but important differences within populations ($P \leq 0.0001$). Moreover, the MDS plot performed with haplotype R_{ST} showed an important Amerindian influence in the Southern populations from Mexico. The first component sets apart the Yucatan population from Chiapas and CVM populations. Having said that, the second component separates the Mestizo populations from the Amerindian populations (Maya, Nahua, Mixteca and Otomi). Of note is that the CVM populations were related to North American populations (Tarahumaras and Amerindians from the North) as well as Nahuas from Xochimilco. This relation could be associated to a linguistic family, where Tarahumaras and Nahuas (contemporary Aztecs) belong to the Uto-Aztecan family. In addition, these findings also show the genetic complexity of Yucatan and Chiapas (Figure 4). Interestingly, the CVM population presented important differences with Chiapas and Yucatan populations (R_{ST}=0.476, $P \leq 0.000$ and R_{ST} =0.536, $P \leq 0.000$, respectively), although it maintains a relationship with the Amerindian lineages.

Finally, in order to determine the relationship between the Mestizo population and the historical parental populations (Amerindian, European and African), we compared overall Mexican Mestizo data from various regions in Mexico using 15 Y-STRs. Furthermore, we also include Middle East populations such as Jew Ashkenazi, Northern Israel and Berbers (Supplementary Information, Table S1) (Figure 4). The MDS plot depicts the complex genetic structure on the Mestizo population. Thus, the CVM population showed a close relation to Berbers and Amerindians, as well as to European populations. On the other hand, Chiapas shared genetic characteristics with Middle East and African populations. With regard to North-Central and Western populations, it showed a close relation to European and Middle East populations. Finally, the Yucatan population presented an important influence of Amerindian and African populations.

Forensic parameters

Forensic parameters using extended haplotype (17 Y-STRs) are shown in Table 2. The 17 Y-STRs showed high discrimination capacity (99.6%) with low random match probability indicating that these loci are useful genetic markers for forensic personal identification and paternity testing in CVM populations. Nevertheless, when the minimal haplotype was used, the discrimination capacity decreased to 79.6%. This finding suggests that in any forensic casework, in which chromosome Y-STRs are used, the extended haplotype should be used in order to decrease the possibility of a false conclusion.

Discussion

As outlined in the introduction, Mexican Mestizo population shows significant ancestral heterogeneity among different geographic regions of Mexico (Luna-Vazquez et al. 2008, Salazar-Flores et al. 2010). This heterogeneity is a consequence of the miscegenation caused mainly by European men with Amerindian women. However, African slaves introduced by the Spaniards also contributed to the Mexican genetic diversity (Beezley 2011). Therefore, the most diversity is found in paternal lineages, whereas the matrilineal ancestry is primarily Amerindian (93%) (Guardado-Estrada et al. 2009, Martinez-Cortes et al. 2012).

Our results indicate that the CVM population is a cosmopolitan population where the Amerindian and European ancestries are the most prominent. European ancestry is related, principally, to the Mediterranean region of Andalucia from where the Spaniard males that arrived to Mexico came from. This ancestry is also associated to Basques traders, who predominated in the port cities of New Spain (Beezley 2011). Both groups present high frequencies of the haplogroup R1b, which is the most frequent lineage in the Mexican Mestizo population (Gaibar et al. 2010; Young et al. 2011).

Additionally, it is worth mentioning that the Andalucian region, invaded by the Arabians and North Africans in the eighth century, contributed to the presence of lineages from African and Middle East in Mexico (Ambrosio et al. 2010; Beezley 2011). In this sense, the Middle East lineages could be associated to subsequent migrations of Crypto-Jews that escaped from Spain to the new colonies (Adams et al. 2008). In contrast, the African lineages increased due to America's miscegenation through of the slave trade (Beezley 2011). Nevertheless, both lineages show heterogeneous patterns and their contribution to the diversity of contemporary Mexican Mestizo population is scarce (Moreno-Estrada et al. 2014).

Focusing on the Amerindian legacy, it showed an interesting population structure, which was represented by the sub-lineages QL54 and QM3, where this last one was the most prominent (77%). Within each sub-lineage high degree of diversity was found, with branches in the network analysis with as much as 10 mutations. This suggests that the parental populations from which the sub-lineages arrived have suffered diverse demographic events. This is coherent with the known history of the Spaniard conquest of Mesoamerica when important demographic changes occurred, but this could also be a reflect of earlier phenomena occurred after the beginning of the peopling of the Americas through the Bering Strait until the Pre-Columbian era (Beezley 2011; Schurr et al. 2012). Both lineages showed an Asian origin with coalesce times to QL54 and QM3 of 7.33 ± 1.038 kya and 13.87 ± 2.148 kya, respectively. These findings are in agreement with previous reports (Battaglia et al, 2013; Sandoval et al. 2012; Schurr et al. 2012). Furthermore, the highest frequency of QM3 in relation to QL54 is congruent, given that QM3 exhibits a clinal distribution southward (Luna-Vazquez et al. 2008; Martinez-Cortes et al. 2012; Salazar-Flores et al. 2010). Hence, the present-day Mexican Mestizo population shows diverse genetic patterns, where even the Amerindian component presents a complex genetic architecture (Moreno-Estrada et al. 2014).

Insofar as the other Mexican populations such as Chiapas, Guanajuato, Jalisco, Yucatan and CVM are concerned, an important difference was detected between Central/Western Mestizo populations and Southern populations ($P \leq 0.001$). This result points to the fact that Mestizo populations from different Mexican regions present different ancestral proportions, which are related to the demographic events that gave origin to each population. Thus, the north-center and western populations such as Guanajuato and Jalisco showed higher European ancestry than southern populations. This distribution is congruent with other reports, which point out that European ancestry shows an increase northward of the country (Luna-Vazquez et al. 2008; Martinez-Cortes et al. 2012; Salazar-Flores et al. 2010). Otherwise, the southern populations such as Chiapas and Yucatan showed highest influence from the Amerindian legacy, which follows a clinal distribution southward (Moreno-Estrada et al. 2014). However, these populations presented complex ancestral patterns. These genetic differences may be related to migratory waves during and after the Spanish colonization (1521–1821) (Beezley 2011). In this regard, historical records reveal an important European (Spaniard-12%, Netherlands/Italy-1%) contribution found in Chiapas and Yucatán (González 2010). These migrations were followed by more European settlements (Germany, French, Italian, Greek, Belgian, Swiss, English, and Russian, among others) during the 19th Century (González 2010). In addition, migratory waves from Asia (China and Japan), Canada (Mennonite from

Manitoba), and even from Oceania (Polynesia) and the Middle East (Lebanon), were also recorded in the 19th and 20th Centuries (González 2010). Similarly, the caste system ("blood purity"), developed by the Spaniards during the colonization, caused a boost of European ancestry in detriment of Amerindian lineages (Beezley 2011). Nevertheless, as attested by a recent report, both Yucatan as well as Chiapas presented a "Mayan component", supporting the fact that even the present-day populations maintain the legacy of local native populations (Moreno-Estrada et al. 2014). Interestingly, these southern populations as well as the CVM population also show some relationship with the African legacy. These findings are in agreement with previous reports in present-day Mexican Mestizo populations, which support the three-hybrid genetic model where the African ancestry is the least frequent (Ge et al. 2010; Coble et al. 2013). Nonetheless, deeper genetic studies should be carried out given that 15 Y-STRs are not enough to support this finding.

Regarding forensic applications, the profound heterogeneity found in the Mexican Mestizo population supports the fact that extended haplotype databases are required. Nevertheless, it would be desirable to increase the number of Y-STRs markers, given that Mexican Mestizos are a young population with small effective population sizes, consequently paternal relatives can share the same profile (Butler 2011, Johnson et al. 2011). Hence, the minimal haplotype analysis may induce mistakes in exclusion and inclusion parameters.

In conclusion, our findings illustrate the complex genetic background of the Mexican Mestizo population and reinforce the need to encompass more geographic regions to generate more robust data. Although the studied sample were Mexican Mestizos, the bearers of Amerindian legacy were remarkable. Moreover, it is worth mentioning that each sub-lineage, within Amerindian ancestry, shows different sub-clades despite the small number of samples. This evidence suggests that further studies in Amerindian populations could be useful in order to elucidate the interesting genetic architecture within the Amerindian lineages. Given that many aspects of the peopling of the Americas are still unsolved, these analyses could contribute, notably, to reconstruct population dynamics. In addition, shedding light on Mexican Amerindian diversity could clarify the migrations that contributed to the peopling of the Americas, where Mexico played a critical role due to the high diversity even within the Mestizo population. About forensic applications, our results provide more information to strengthen male genetic discrimination in kin relationships. However, further research is needed to analyze the contributions of other demographic events in the genetic wealth of this complex population. To our knowledge, this report constitutes the first study that describes the genetic substructure of Amerindian lineages in Mexican Mestizos.

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Declaration of Interests

The authors declare that they have no conflict of interests and no financial relationship with the organization sponsoring the research.

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Table 1

Title: Allelic frequencies with 17 Y-STRs loci in the Mexican Mestizo population from Central Valley of Mexico.

	GUANAJUATO (n=63)																
Allele/n	**DYS19**	**DYS385a**	**DYS385b**	**DYS389I**	**DYS389II**	**DYS390**	**DYS391**	**DYS392**	**DYS393**	**DYS437**	**DYS438**	**DYS439**	**DYS448**	**DYS456**	**DYS458**	**DYS635**	**YGATAH4**
6																	
7																	
8							0.063										
9							0.016				0.063						
10		0.111					0.286		0.032		0.19	0.048					
11		**0.381**					**0.508**	**0.254**	0.127		0.206	0.143					**0.444**
12	0.016	0.111		0.111			0.048	0.016	0.111		**0.524**	**0.524**					**0.46**
13	0.254	0.095	0.048	**0.714**			0.016	0.413	**0.651**		0.016	0.175		0.016			0.095
14	**0.54**	0.143	**0.381**	0.175			0.048	**0.238**	0.079	**0.476**		0.111		0.032	0.048		
15	0.111	0.079	0.159				0.016	0.048		**0.492**				**0.444**	0.095		
16	0.048	0.032	0.095					0.032		0.016				0.349	0.238		
17	0.032	0.016	0.111							0.016				0.143	**0.508**		
18		0.032	0.111										0.079	0.016	0.111		
19			0.048										**0.476**				

20													0.365			0.079	
21			0.048			0.016							0.079			0.063	
22						0.079										0.286	
23						0.206										**0.476**	
24						**0.603**										0.095	
25						0.079											
26					0.016	0.016											
27																	
28					0.048												
29					**0.444**												
30					**0.413**												
31					0.079												
32																	
33																	
k	6	9	8	3	5	6	8	6	5	4	5	5	4	6	5	5	3

	PUEBLA (n=47)																
Allele/n	**DYS19**	**DYS385a**	**DYS385b**	**DYS389I**	**DYS389II**	**DYS390**	**DYS391**	**DYS392**	**DYS393**	**DYS437**	**DYS438**	**DYS439**	**DYS448**	**DYS456**	**DYS458**	**DYS635**	**YGATAH4**
6											0.021						
7																	

8																	
9		0.021					0.021				0.106						0.021
10		0.064					**0.532**		0.021		0.128	0.043					
11		**0.277**					0.362	0.17	0.128		0.277	**0.319**					0.277
12	0.064	0.128	0.021	0.106			0.064	0.064	0.234		**0.468**	**0.383**					**0.574**
13	**0.383**	0.085	0.128	**0.702**				**0.404**	**0.532**			0.255		0.021			0.128
14	**0.34**	0.191	**0.213**	0.191			0.021	0.191	0.085	**0.532**				0.043	0.021		
15	0.191	0.17	0.106				0	0.043		0.404				**0.447**	0.128		
16	0.021	0.064	0.17					0.106		0.064				0.255	0.277		
17		0	0.191					0.021		0			0.021	0.17	**0.34**		
18			0.106										0.043	0.043	0.128		
19			0.064										**0.447**	0.021	0.064	0.021	
20													0.362		0.043	0.021	
21													0.128			0.128	
22						0.064										0.362	
23						0.277										**0.426**	
24						**0.532**										0.043	
25						0.106											
26						0.021											
27																	
28					0.085												

29					**0.404**												
30					0.383												
31					0.106												
32					0.021												
33																	
k	5	8	8	3	5	5	5	7	5	3	5	4	5	7	7	6	4

	QUERÉTARO (n=120)																
Allele/n	**DYS19**	**DYS385a**	**DYS385b**	**DYS389I**	**DYS389II**	**DYS390**	**DYS391**	**DYS392**	**DYS393**	**DYS437**	**DYS438**	**DYS439**	**DYS448**	**DYS456**	**DYS458**	**DYS635**	**YGATAH4**
6																	
7											0.008						
8							0.025										
9		0.017					0.017				0.083						0.008
10		0.099	0.008				**0.504**		0.033		0.281	0.058					0.05
11		**0.331**	0.025				0.405	**0.339**	0.083		0.248	0.298					0.413
12	0.017	0.083	0.033	0.149			0.033	0.033	0.157		**0.331**	**0.413**					0.413
13	0.256	0.124	0.083	**0.678**			0.008	**0.339**	**0.612**	0.008	0.05	0.207		0.008			0.116
14	**0.471**	0.141	**0.273**	0.165			0.008	0.182	0.099	**0.529**		0.017		0.083	0.017		
15	0.174	0.116	0.157	0.008				0.041	0.017	0.397		0.008		**0.479**	0.066		
16	0.041	0.058	0.083					0.058		0.066			0.008	0.264	**0.339**		

17	0.041	0.033	0.124					0.008					0.008	0.132	**0.372**		
18			0.107										0.099	0.025	0.165		
19			0.058										**0.397**	0.008	0.041		
20													0.289			0.066	
21			0.05			0.05							0.132			0.198	
22						0.107							0.058			0.231	
23						0.248										**0.43**	
24						**0.529**							0.008			0.066	
25						0.058										0.008	
26																	
27					0.008	0.008											
28					0.149												
29					**0.372**												
30					**0.355**												
31					0.066												
32					0.041												
33					0.008												
k	6	9	11	4	7	6	7	7	6	4	6	6	8	7	6	6	5

k= Number of alleles. Bold numbers represent more frequent alleles.

Table 2

Title: Forensic statistic parameters in Central Valley of Mexico populations.

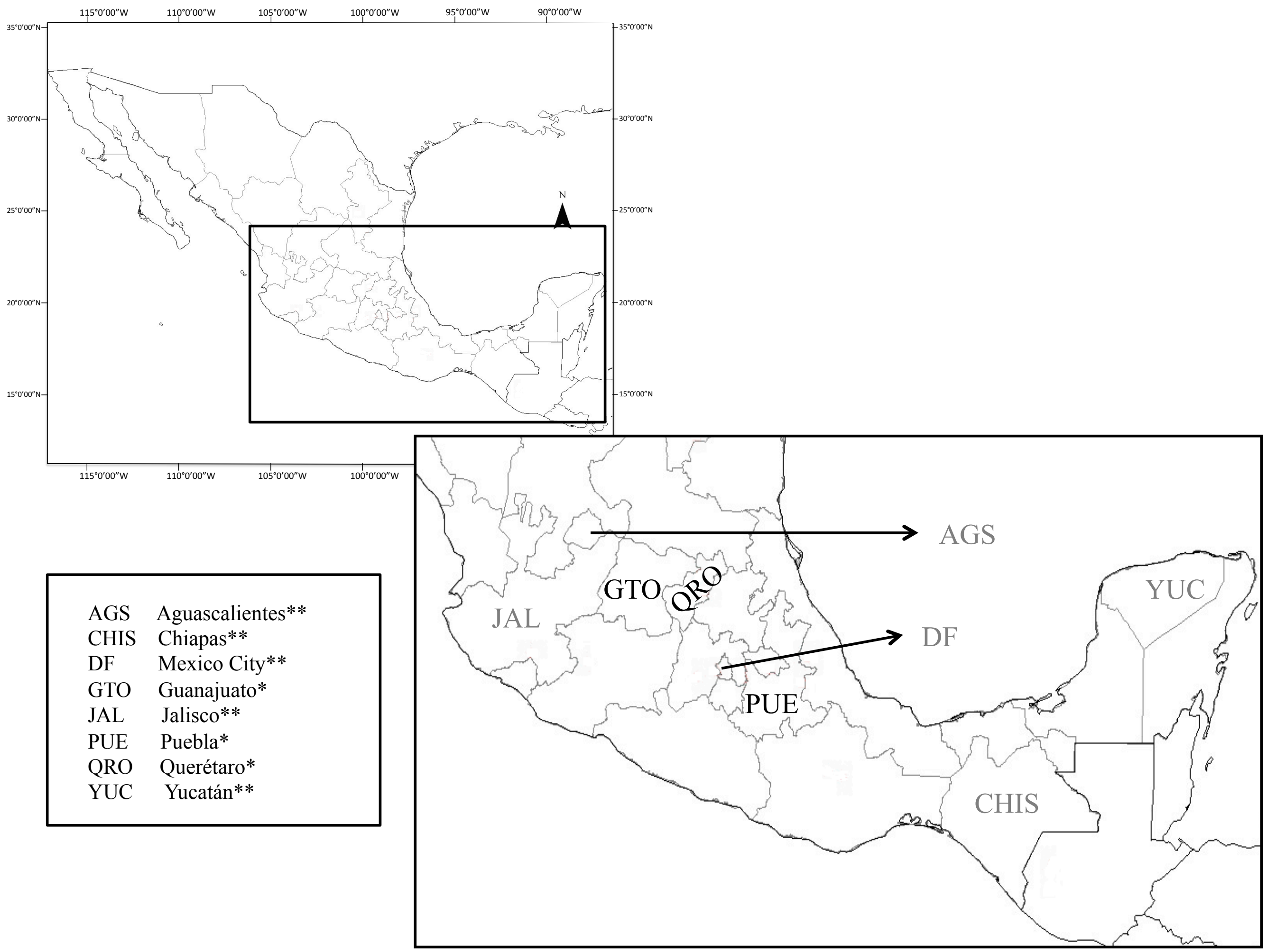
Footnote: pD=power of discrimination, MP=match probability, DC= discrimination capacity, n= sample size, nuh=number of unique haplotypes

	LOCUS	Whole	GTO	PUE	QRO
pD	DYS19	0.680	0.628	0.696	0.679
	DYS385a	0.821	0.792	0.826	0.821
	DYS385b	0.845	0.789	0.846	0.853
	DYS389I	0.475	0.447	0.459	0.491
	DYS389II	0.684	0.623	0.671	0.707
	DYS390	0.627	0.580	0.625	0.641
	DYS391	0.616	0.651	0.581	0.580
	DYS392	0.739	0.705	0.753	0.731
	DYS393	0.589	0.541	0.638	0.583
	DYS437	0.554	0.531	0.550	0.558
	DYS438	0.717	0.642	0.676	0.741

	DYS439	0.670	0.660	0.684	0.694
	DYS448	0.692	0.627	0.651	0.728
	DYS456	0.682	0.659	0.702	0.675
	DYS458	0.722	0.662	0.769	0.713
	DYS635	0.703	0.672	0.669	0.714
	YGATAH4	0.623	0.582	0.577	0.643
MP		0.003	0.003	0.004	0.001
DC (%)		99.57	100	100	100
n		231	63	47	121
nuh		230	63	47	121

Figure 1.

Title: Map of the Mexican Republic and locations of sampling and comparison.

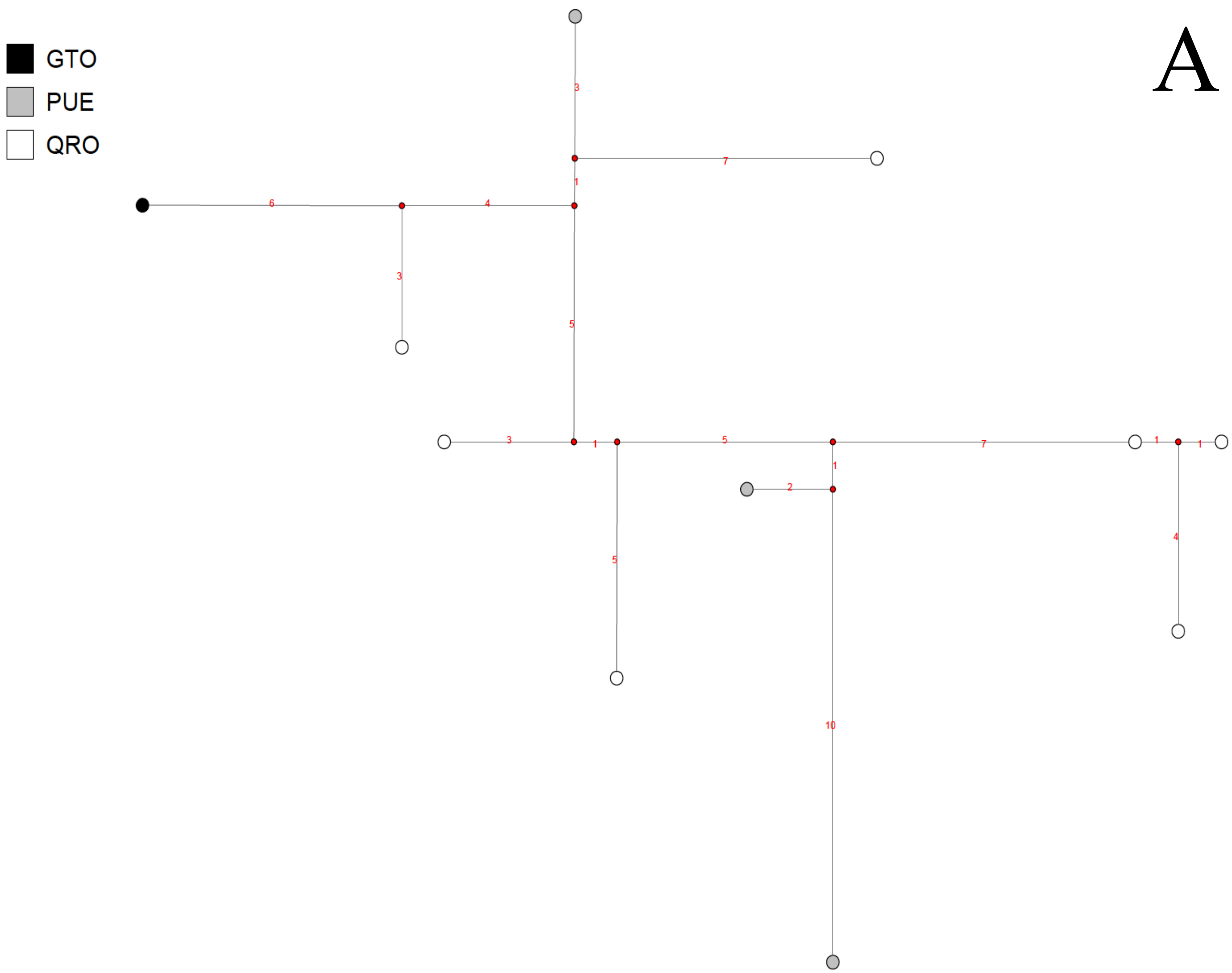


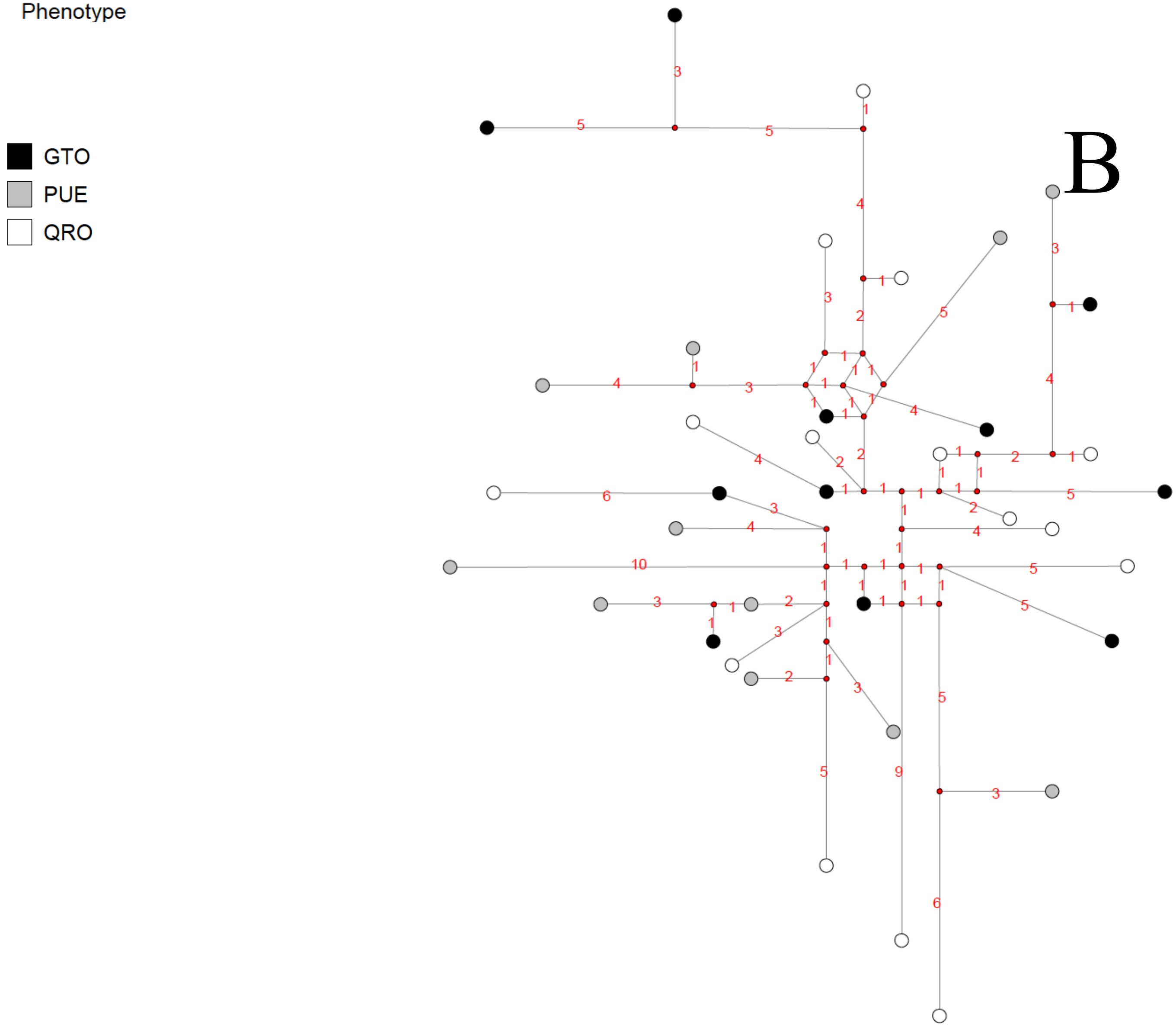
* This study.

** Populations for comparison.

Figure 2.

Title: Median-Joining network of ancestral Amerindian lineages in Mestizo population from Mexico using 15 Y-STRs. A) Sub-lineage QL54. B) Sub-lineage QM3. The node size reflects the number of individuals having the same haplotype.

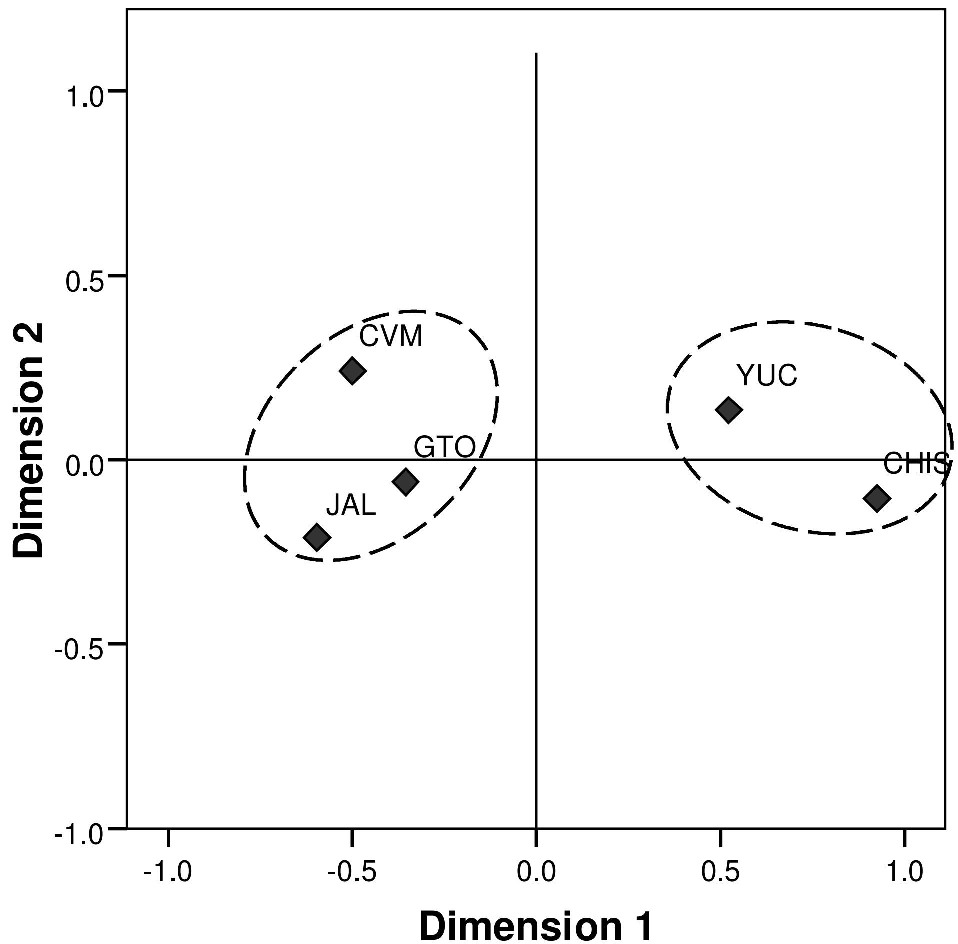




GTO: Guanajuato, PUE: Puebla, QRO: Querétaro. The numbers indicate the number of mutations.

Figure 3.

Title: MDS plot of haplotypes R_{ST} pairwise differences using 15 Y-STRs.



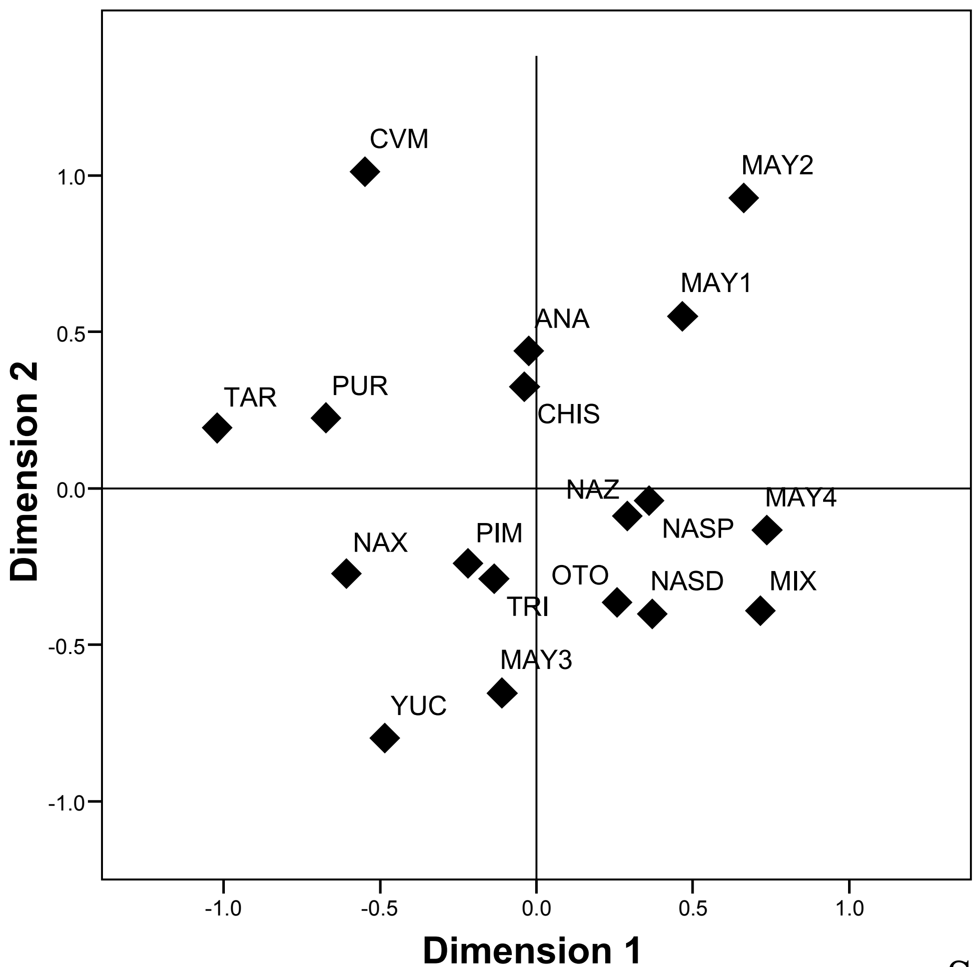
Stress=0.0217

CMV=Central Valley of Mexico, CHIS=Chiapas, GTO=Guanajuato, JAL=Jalisco, YUC=Yucatan.

Figure 4.

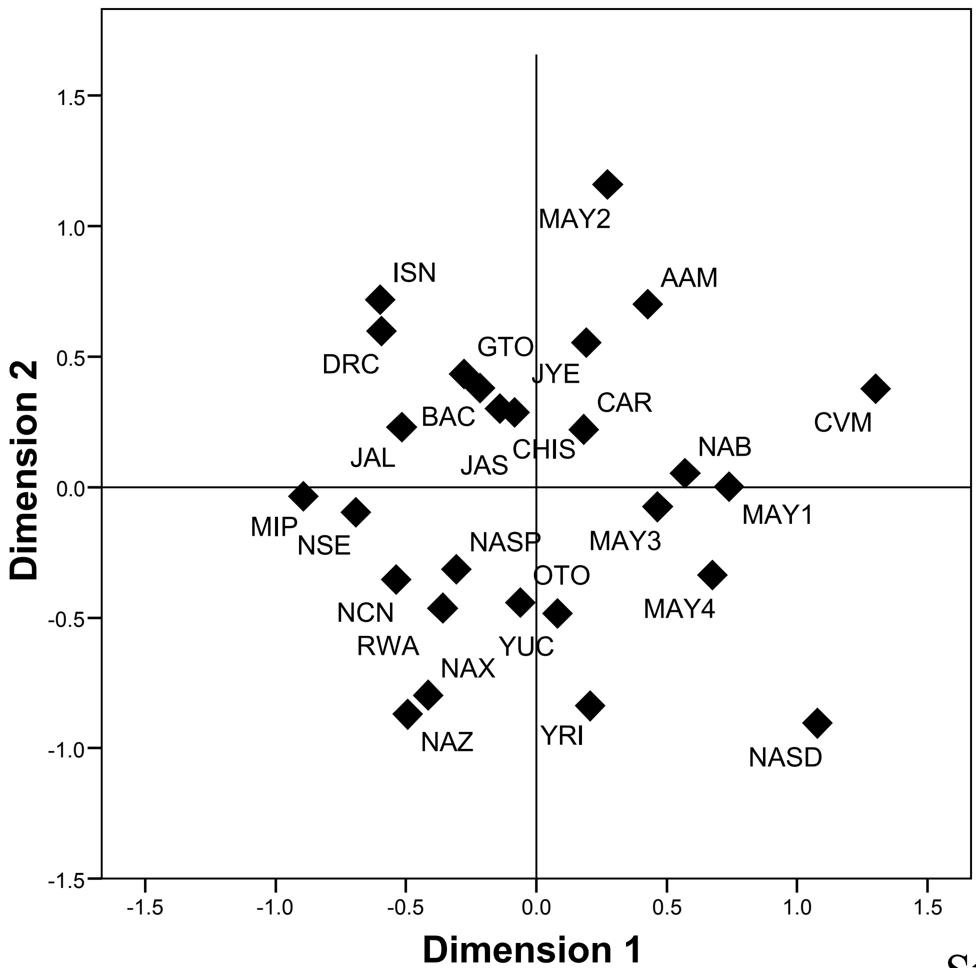
Title: MDS plot of haplotypes R_{ST} pairwise differences using 15 Y-STRs. A). Comparison between Amerindians and Mestizo populations. B) Comparison between Mestizo and parental populations (Amerindians, European and Africans).

A



Stress: 0.06119

B



Stress: 0.06983

AAM=African American, ANA=Ameridians from North America, BAC= Basque Country, CAR=Central Africa Republic, CMV=Central Valley of Mexico, CHIS=Chiapas, DRC=Democratic Republic of the Congo, ISN=Northern Israel, JAS=Jews Ashkenazi, JYE=Jews Yemenite, MAY1=Mayas from Cakchikel, Guatemala, MAY2=Mayas from Yucatan, MAY3=Mayas from

Campeche, MAY4=Mayas from Yucatan, MIP=Mediterranean Iberian Peninsula, MIX=Mixtecas, NAB=North Africa Berbers, NASD=Nahuas from Santo Domingo Atocpan, NASP=Nahuas from San Pedro Atocpan, NAX=Nahuas from Xochimilco, NAZ=Nahuas from Zitlala, NCN=North Central Africa, NSE=Southeastern Nigeria, OTO=Otomis, PIM=Pimas, PUR=P'urhepechas, RWA=Rwanda, TAR=Tarahumaras, TRI=Triquis, YRI=Yorubas from Nigeria, YUC=Yucatan.

Supplementary Information

Table S1

Title: Populations using for comparison purposes

Population	Origin	n	Key	Number of Y-STR	Analysis	Reference
Mexican Mestizo	Guanajuato	63	CVM	17	MDS/Network	This study
	Puebla	47				
	Querétaro	121				
	Yucatán	170	YUC	17	MDS	Salazar-Flores J et al.; 2010
	Chiapas	170	CHIS			
	Jalisco	185	JAL			
	Guanajuato	168	GTO	12		

	Aguascalientes	293	AGS			
	Mexico City	357	DF	11	MDS	Luna-Vazquez A et al.; 2008
Amerindian Mexico	Mayas-Cakchikel	43	MAY1	15	MDS	Regueiro M et al.; 2013
	Mayas-Yucatan	72	MAY2			
	Mayas-Campeche	11	MAY3	15	MDS	Xu H et al.; 2014
	Mayas-Yucatan	19	MAY4	17	MDS	Sandoval K et al.; 2012
	Mixtecs	2	MIX			
	Nahuas San Pedro Atocpan	7	NASP			
	Nahuas Santo Domingo	15	NASD			

	Ocotitlan						
	Nahuas Xochimilco	15	NAX				
	Nahuas Zitlala	19	NAZ				
	Otomis	4	OTO				
	Pimas	49	PIM				
	P'urhepecha	6	PUR				
	Tarahumaras	13	TAR				
	Triquis	22	TRI				
	Gwich'in	7					
	Inuvialuit	6					
Amerindian North America	Tlingit	7	ANA	15		Network	Dulik M C et al.; 2012
	Mi'kmaq	1					
	Wiyot	1					

	Canadian Metis	1				
Asia	Tuva Republic	23	TUV	15	Network	Regueiro M et al.; 2013
	Northeast tip of Siberia	7	NES			
Africa	Bagandu, Central Africa Republic (Biaka Pygmy)	33	CAR	15	MDS	Xu H et al.; 2014
	Ituri Forest, Democratic Republic of the Congo (Mbuti Pygmies)	13	DRC			
	Nigeria	35	YRI			

(Yoruba)		
Zaria, North Central Nigeria (Hausa)	16	NCN
Northern Israel (Druze)	40	ISN
South Africa (Ashkenazi Jews)	18	JAS
Enugu, Southeastern Nigeria (Ibo)	7	NSE
Yemen (Yemenite Jews)	21	JYE

	Rwanda	67	RWA	15	MDS	Balamurugan K et al.; 2012
	North Africa Berbers	81	NAB	15	MDS	Gaibar M et al.; 2011
African American	United States of America	12	AAM	15	MDS	Xu H et al.; 2014
European	Mediterranean Iberian Peninsula	49	MIP	15	MDS	Gaibar M et al.; 2010
	Basque Country	182	BAC	15	MDS	Valverde L et al.; 2012.

Table S2

Title: Y-STR Mexican Mestizo haplotypes, haplogroup identification from haplotype definition, fitness score and probability.

Haplotype	DYS19	DYS385a	DYS385b	DYS389I	DYS389II	DYS390	DYS391	DYS392	DYS393	DYS437	DYS438	DYS439	DYS448	DYS456	DYS458	DYS635	YGATAH4	Haplogroup	Fitness Score	Probability	Guanajuato	Puebla	Querétaro	Total
H1	12	10	13	13	29	23	11	13	13	15	12	13	19	15	17	23	13	R1b	36	1	1			1
H2	12	11	13	13	30	24	11	13	13	15	13	13	19	15	17	23	12	R1b	38	1			1	1
H3	12	11	13	14	30	23	11	14	14	14	10	10	19	14	17	21	12	N	66	1		1		1
H4	12	12	16	13	29	23	10	12	12	15	9	11	20	15	14	19	12	J2a1b	23	0.97		1		1
H5	12	12	16	13	29	23	11	14	12	14	9	12	21	19	16	22	12	T	16	0.65		1		1
H6	12	13	15	13	30	24	10	14	11	14	12	13	19	17	15	25	13	Q	21	0.94			1	1
H7	13	10	12	14	31	24	11	13	13	15	12	13	21	15	16	23	12	R1b	23	0.99		1		1
H8	13	10	13	12	28	23	10	14	11	14	12	12	18	16	18	23	11	Q	28	0.64			1	1
H9	13	10	13	12	28	24	11	13	12	15	12	11	19	15	16	23	12	R1b	33	1		1		1
H10	13	10	13	13	29	24	11	13	12	15	12	12	19	17	17	23	12	R1b	39	1	1			1
H11	13	10	13	13	29	25	14	14	13	15	12	14	19	15	17	23	12	R1b	21	1	1			1
H12	13	10	13	14	30	24	11	13	13	14	12	12	18	15	15	23	11	R1b	35	1			1	1
H13	13	10	14	13	29	24	11	13	13	15	12	12	19	16	16	23	12	R1b	53	1			1	1
H14	13	10	14	14	30	24	11	13	13	14	12	13	19	16	17	23	11	R1b	47	1		1		1

H15	13	10	15	13	31	24	11	14	13	16	12	12	20	15	17	24	12	R1b	23	0.99	1			1
H16	13	11	14	13	30	25	14	13	12	15	12	12	19	15	17	23	13	R1b	26	1			1	1
H17	13	11	16	13	30	24	10	11	13	14	10	11	20	17	16	22	12	E1b1b	51	0.99	1			1
H18	13	11	16	13	31	24	10	15	11	14	10	11	19	17	19	22	11	Q	34	0.99		1		1
H19	13	11	21	13	29	23	10	11	12	14	10	13	20	15	19	20	13	J1	20	0.96			1	1
H20	13	12	13	14	29	23	10	13	10	14	9	11	20	15	17	22	11	T	23	0.74			1	1
H21	13	12	15	13	29	24	11	13	13	15	12	12	19	16	16	23	11	R1b	83	1	1			1
H22	13	12	18	13	30	23	10	11	13	14	12	12	20	16	17	22	12	E1b1b	48	0.96	1			1
H23	13	13	14	14	30	24	8	11	11	14	10	10	20	15	18	21	12	E1b1b	25	0.99	1			1
H24	13	13	14	14	30	24	10	11	13	14	10	10	20	16	18	21	12	E1b1b	52	1			1	1
H25	13	13	15	13	29	24	10	14	12	14	11	12	20	15	20	23	11	Q	44	0.99		1		1
H26	13	13	16	13	29	23	8	11	12	15	9	12	20	16	17	22	12	J2a1b	31	0.48	1			1
H27	13	13	17	13	29	23	11	11	12	15	9	11	16	15	16	21	12	T	28	0.84			1	1
H28	13	13	17	13	30	24	10	14	13	14	11	12	22	16	16	22	11	Q	73	1			1	1
H29	13	13	18	13	30	24	10	14	13	14	11	13	20	15	17	22	11	Q	76	1	1			1
H30	13	14	15	13	30	21	11	11	13	14	11	11	19	15	16	24	13	E1b1a	30	0.98			1	1
H31	13	14	16	13	29	24	13	15	11	14	12	14	19	17	16	24	13	Q	13	0.98	1			1
H32	13	14	16	13	30	23	11	16	12	14	11	11	20	15	17	22	12	Q	47	1			1	1
H33	13	14	17	12	29	22	11	16	13	14	11	10	20	15	16	22	11	Q	47	1			1	1
H34	13	14	17	13	30	24	11	14	13	14	11	12	20	16	16	22	11	Q	73	1	1			1
H35	13	14	17	14	31	24	10	14	13	14	9	13	19	15	15	21	11	T	58	0.99	1			1
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H40	13	14	18	13	29	24	10	16	13	13	10	11	20	15	16	22	11	Q	45	0.99			1	1

H41	13	14	18	13	30	23	10	14	11	14	12	11	20	17	16	22	11	Q	49	1		1		1
H42	13	14	18	13	30	24	11	15	13	14	6	11	20	16	17	22	12	Q	25	0.97		1		1
H43	13	14	18	13	32	24	10	17	10	14	10	12	20	18	17	22	11	Q	16	0.53			1	1
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H45	13	14	18	14	31	26	10	14	14	14	11	11	19	16	16	22	11	Q	52	1	1			1
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H56	13	15	17	14	31	24	10	16	13	14	11	11	20	16	15	23	12	Q	45	0.98		1		1
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H70	13	17	18	13	28	24	8	11	14	14	10	12	18	15	16	22	11	E1b1b	32	0.99			1	1
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H190	15	11	15	14	29	24	12	13	13	15	12	11	17	15	16	23	12	R1b	31	1		1		1
H191	15	11	15	14	30	24	10	13	13	14	12	11	18	15	18	23	11	R1b	49	1		1		1
H192	15	11	16	13	30	22	11	16	13	14	9	11	19	15	15	21	11	T	38	1			1	1
H193	15	12	13	13	30	24	11	13	13	15	12	11	20	15	17	23	12	R1b	44	1			1	1
H194	15	12	14	13	29	24	11	13	13	15	12	11	19	17	17	23	12	R1b	60	1			1	1
H195	15	12	15	12	29	24	10	14	12	14	12	14	19	16	17	23	13	E1b1b	88	1	1		1	2

H196	15	12	16	13	28	23	8	11	14	15	11	12	20	15	15	22	12	J2a1 x J2a1-bh	21	0.4	1			1
H197	15	12	19	13	29	22	10	11	13	14	10	11	20	15	18	21	10	J1	52	0.99			1	1
H198	15	13	14	13	29	22	10	11	14	16	10	13	21	15	19	22	13	G2a	46	1			1	1
H199	15	13	14	13	29	23	10	13	13	14	12	13	19	17	18	22	12	Q	47	0.96	1			1
H200	15	13	14	13	29	24	8	11	13	15	11	11	20	15	17	24	12	R1a	24	0.82			1	1
H201	15	13	15	12	29	21	10	11	14	16	10	11	22	15	16	21	11	G2a	76	1			1	1
H202	15	13	15	12	30	24	10	11	14	16	10	11	20	14	17	21	11	G2a	52	0.98			1	1
H203	15	13	16	12	29	23	10	11	14	16	10	13	21	17	15	21	12	G2a	48	1		1		1
H204	15	13	17	12	28	23	10	11	12	14	9	11	21	15	18	22	11	J2a1 x J2a1-bh	60	0.51			1	1
H205	15	14	15	12	27	23	10	14	13	14	9	11	19	15	16	21	10	T	74	1			1	1
H206	15	14	17	13	30	24	12	13	13	14	11	11	21	15	18	22	12	Q	38	0.98			1	1
H207	15	14	18	13	29	22	10	11	12	16	9	13	19	13	16	22	11	J2b	50	1		1		1
H208	15	14	18	13	29	23	11	13	14	15	11	11	19	16	18	22	11	Q	46	0.93	1			1
H209	15	15	16	13	29	23	11	12	14	15	11	11	20	15	15	20	12	I2b1	40	1		1		1
H210	15	15	17	12	28	23	11	11	12	14	9	12	20	14	16	21	10	H	50	0.51			1	1
H211	15	15	17	12	30	23	10	11	14	15	10	12	21	14	17	22	11	G2a	41	0.96	1			1
H212	15	15	17	13	30	23	10	12	11	14	12	13	20	16	19	21	11	E1b1a	27	0.72		1		1
H213	15	15	17	13	30	23	10	14	13	14	11	12	19	16	16	23	10	Q	61	0.99			1	1
H214	15	15	18	14	32	23	10	11	14	14	11	12	20	15	15	21	11	E1b1a	54	0.71			1	1
H215	16	10	14	13	30	24	11	11	11	14	11	10	20	16	15	23	12	R1a	34	1	1			1
H216	16	11	12	15	30	23	10	11	13	15	11	12	22	15	17	22	11	I2a1	38	1			1	1
H217	16	11	14	13	31	27	11	11	13	14	11	10	20	16	15	24	13	R1a	51	1			1	1
H218	16	11	15	13	30	24	11	13	13	14	12	12	18	15	16	23	11	R1b	45	0.99	1			1
H219	16	12	14	13	28	24	11	13	13	15	12	11	19	15	17	24	13	R1b	34	1			1	1

H220	16	12	14	13	29	24	11	14	13	15	12	12	19	15	17	23	12	R1b	49	1		1		1
H221	16	13	18	12	28	24	11	11	13	15	10	12	20	15	17	23	11	I2a(xI2a1)	55	0.97			1	1
H222	16	14	16	12	26	24	12	14	11	17	11	14	19	17	17	22	12	L	15	0.99	1			1
H223	16	17	19	13	29	21	10	11	13	14	11	11	21	16	16	21	11	E1b1a	67	1			1	1
H224	17	11	12	13	28	23	10	11	13	15	10	11	21	14	16	21	11	I2a1	85	1			1	1
H225	17	11	12	13	28	24	10	11	13	14	10	13	21	14	16	22	11	I2a1	63	1			1	1
H226	17	12	14	13	28	22	9	11	12	15	10	12	21	14	17	21	12	I2a1	45	1			1	1
H227	17	13	14	13	28	23	9	11	14	14	10	13	21	13	14	23	11	I2a1	21	0.99	1			1
H228	17	14	12	13	28	22	12	11	10	15	10	12	21	14	17	21	12	I2a(xI2a1)	14	0.52			1	1
H229	17	17	18	13	30	21	11	11	13	14	11	11	21	15	17	20	13	E1b1a	55	1	1			1
H230	17	17	21	14	31	21	10	11	15	14	11	12	21	17	18	21	11	E1b1a	48	1			1	1
Total																								231

Table S3.

Title: The most frequent haplotypic combinations found in CVM population.

DYS19	DYS385a	DYS385b	DYS389I	DYS389II	DYS390	DYS391	DYS392	DYS393	DYS437	n
13	16	17	13	30						
13	16	17	13	30						3
13	16	17	13	30						
14	10	11	13	29						2
14	10	11	13	29						
14	11	13	14	30						2
14	11	13	14	30						
15	13	14	13	29						3

15	13	14	13	29			
15	13	14	13	29			
15	14	18	13	29			2
15	14	18	13	29			
12	12	16	13	29	23		2
12	12	16	13	29	23		
13	13	14	14	30	24		2
13	13	14	14	30	24		
13	15	17	13	30	24		
13	15	17	13	30	24		3
13	15	17	13	30	24		
13	15	19	13	30	24		2
13	15	19	13	30	24		
14	10	14	13	30	24		2
14	10	14	13	30	24		
15	15	17	13	30	23	10	2

15	15	17	13	30	23	10		
14	11	13	13	29	24	11	13	2
14	11	13	13	29	24	11	13	
14	11	14	13	29	24	11	13	11
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	11	14	13	29	24	11	13	
14	13	16	13	30	23	10	11	3
14	13	16	13	30	23	10	11	

14	13	16	13	30	23	10	11			
14	11	15	13	29	24	11	13	13	15	
14	11	15	13	29	24	11	13	13	15	3
14	11	15	13	29	24	11	13	13	15	

Table S4.

Title: Allelic combination in DYS385a/b locus found in Central Valley Mexican populations.

DYS385a/b			
Genotype	**GTO**	**PUE**	**QRO**
n	63	47	121
9,10	-	0.021	0.008
9,13	-	-	0.008
10,11	-	-	0.025
10,12	-	0.021	-
10,13	0.048	0.021	0.017
10,14	0.048	0.021	0.042
10,15	0.016	-	0.008
10,16	-	-	0.008
11,12	-	-	0.025
11,13	-	0.043	0.042
11,14	0.286	0.149	0.158

11,15	0.063	0.064	0.075
11,16	0.016	0.021	0.025
11,17	0.016	-	-
11,21	-	-	0.008
12,13	-	0.043	0.017
12,14	-	0.043	0.042
12,15	0.063	-	0.008
12,16	0.016	0.043	-
12,18	0.016	-	-
12,19	-	-	0.008
12,21	0.016	-	0.017
13,14	0.048	-	0.033
13,15	-	0.021	0.033
13,16	0.016	0.064	0.017
13,17	-	-	0.025
13,18	0.016	-	0.017
13,19	0.016	-	-
14,15	0.016	0.021	0.033
14,16	0.048	0.021	0.017
14,17	0.032	0.043	0.042

14,18	0.048	0.064	0.042
14,19	-	0.043	-
14,21	-	-	0.008
15,16	-	0.021	0.025
15,17	0.048	0.128	0.033
15,18	-	0.021	0.033
15,19	0.016	-	0.025
15,21	0.016	-	-
16,17	0.016	0.021	0.025
16,18	0.016	0.021	0.008
16,19	-	0.021	0.017
17,18	0.016	-	0.008
17,19	-	-	0.008
17,21	-	-	0.008
18,19	0.016	-	-
18,21	0.016	-	-

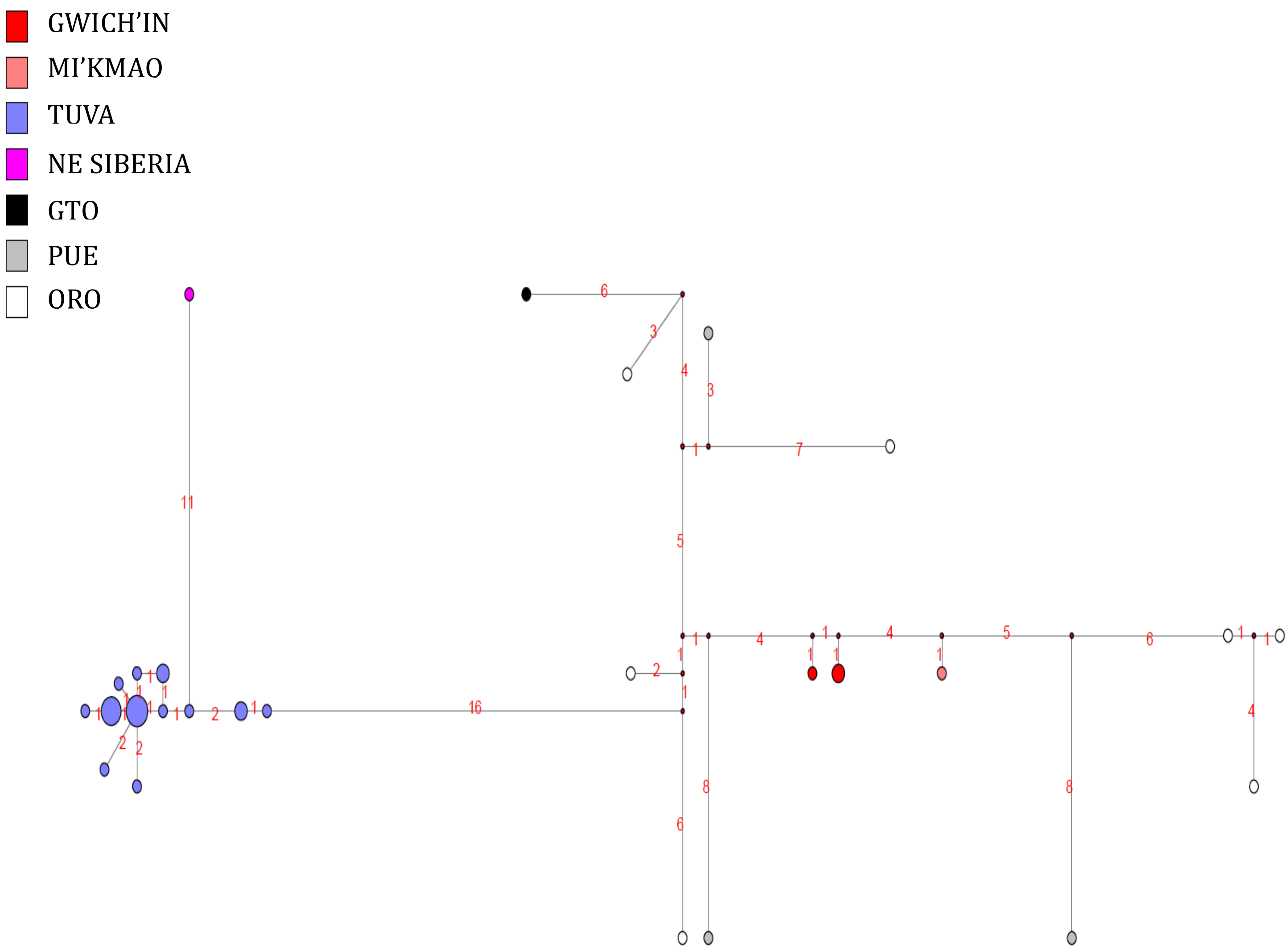
Table S5.

Title: Locus diversity and pairwise differences in the Mestizo populations studied.

Population	**Guanajuato**	**Puebla**	**Querétaro**
# of samples	63	47	120
# of haplotypes	63	47	120
Haplotype diversity	1 ± 0.003	1 ± 0.004	1 ± 0.001
Pairwise differences	11.572 ± 5.314	10.513± 4.878	11.566 ± 5.278

Figure S1.

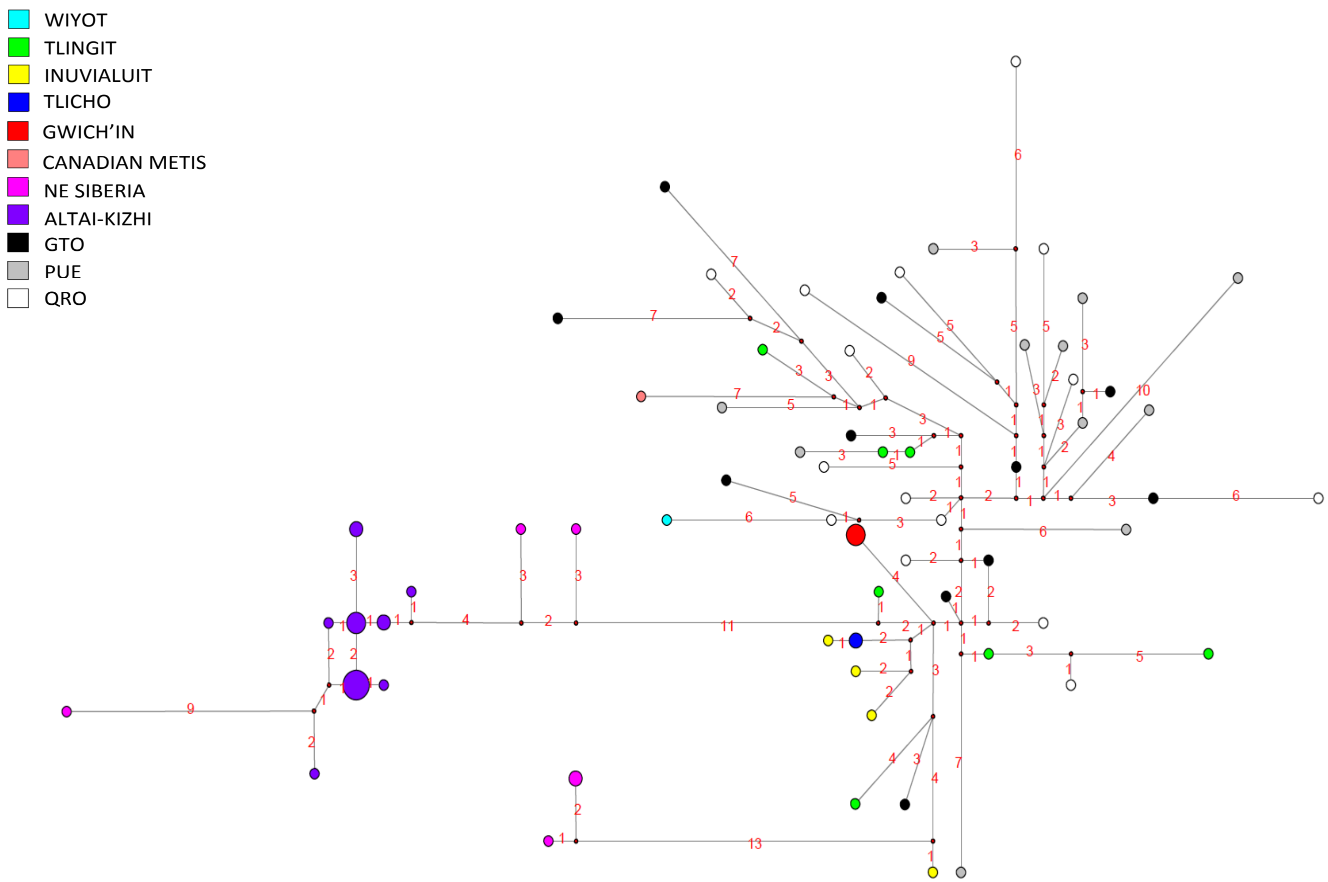
Title: Comparison of QL54 lineage using Median-Joining network with 15 Y-STRs. The node size reflects the number of individuals having the same haplotype.



GTO: Guanajuato, PUE: Puebla, QRO: Querétaro. The numbers indicate the number of mutations.

Figure S2.

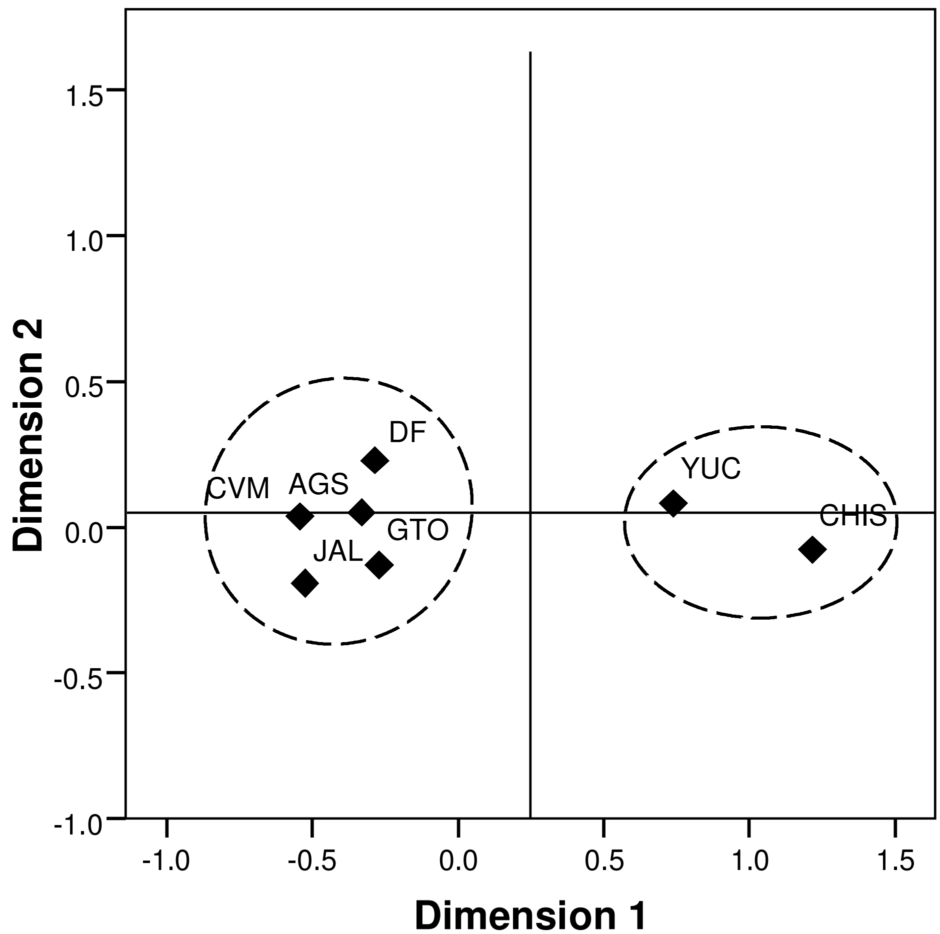
Title: Comparison of QM3 lineage using Median-Joining network with 15 Y-STRs. The node size reflects the number of individuals having the same haplotype.



GTO: Guanajuato, PUE: Puebla, QRO: Querétaro. The numbers indicate the number of mutations.

Figure S3.

Title:MDS plot of haplotypes R_{ST} pairwise differences using the minimal haplotype.



Stress=0.00389

AGS=Aguascalientes, CMV=Central Valley of Mexico, CHIS=Chiapas, DF= Mexico City, GTO=Guanajuato, JAL=Jalisco, YUC=Yucatan.