ELSEVIER

CrossMark

Archives of Medical Research 45 (2014) 409–416

Archives of Medical Research

ORIGINAL ARTICLE

Influence of *CYP1A1*2C* on High Triglyceride Levels in Female Mexican Indigenous Tarahumaras

Claudia E. Bailón-Soto,[a] Carlos Galaviz-Hernández,[a] Blanca P. Lazalde-Ramos,[b] Daniel Hernández-Velázquez,[c] José Salas-Pacheco,[d] Ismael Lares-Assef,[a] and Martha Sosa-Macías[a]

[a]*Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional del IPN Unidad Durango, CIIDIR-IPN, Durango, México*
[b]*Laboratorio de Medicina Molecular, Unidad Académica de Ciencias Químicas, Universidad Autónoma de Zacatecas, México*
[c]*Instituto Nidiac, Durango, México*
[d]*Instituto de Investigación Científica de la UJED, Durango, México*

Received for publication November 8, 2013; accepted May 20, 2014 (ARCMED-D-13-00626).

Background and Aims. High triglyceride levels are closely related to cardiovascular disease. Its development lays on age, diet, physical activity, ethnicity and genetic factors. Among the last, the *CYP1A1*2C* allele has an influence on the metabolism of cholesterol and other fatty acids. We undertook this study to determine the frequency of *CYP1A1*2C* and its association with triglyceride levels in Mexican indigenous Tarahumaras and Tepehuanos.

Methods. Anthropometric and biochemical data were recorded. Genotyping of *CYP1A1*2C* by RT-PCR was done in 110 Tepehuano, 69 Tarahumara and 64 Mestizo.

Results. Significant differences in age, waist diameter, BMI, creatinine, glucose, cholesterol, triglycerides, HDL and VLDL measurements were found between Tarahumaras and Tepehuanos ($p < 0.05$). Additionally, Tarahumara women showed the highest values of waist diameter, BMI and triglycerides ($p < 0.05$). It was found that Tarahumaras showed a significant association between high triglyceride levels and *CYP1A1*2C* allele (OR = 2.57; 95% CI 1.12–5.88, $p = 0.024$) under a recessive inheritance model. However, the Tepehuano group showed a significant protective association between normal triglyceride levels and *CYP1A1*2C* polymorphism (OR = 0.28; 95% CI 0.10–0.80, $p = 0.015$) following a dominant inheritance model. The same pattern was observed after analysis with females of both ethnicities.

Conclusion. A significant association between *CYP1A1*2C* and high triglyceride levels in Amerindian Tarahumaras from Chihuahua has been found; this allele was significantly associated with normal triglyceride levels in Tepehuanos from Durango, Mexico. Further studies are needed to elucidate the genetic role of *CYP1A1* in cardiovascular disease susceptibility.

Key Words: High triglyceride levels, Tarahumaras, Tepehuanos, *CYP1A1*.

Introduction

Because the liver plays a fundamental role in the synthesis and transport of fatty acids, it can be associated with triglyceride disorders. Hypertriglyceridemia is a frequent lipid disorder, which can be defined as an abnormal increase of triglyceride concentration in the blood (1). Primary hypertriglyceridemia has a genetic etiology and is inherited. Secondary hypertriglyceridemia, on the other hand, has underlying causes such as diabetes or alcoholism. A study revealed that primary and secondary hypertriglyceridemia are often associated with other lipid abnormalities (2). High triglyceride levels are compelling factors of atherogenic dyslipidemia and can increase cardiovascular risk (3).

Hypertriglyceridemia, abdominal obesity along with a large waist, genetics, low metabolism, null physical activity, a diet rich in saturated fat and very high caloric content and

Address reprint requests to: Martha Sosa-Macías, Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional del IPN Unidad Durango, CIIDIR-IPN, Sigma 119 Fracc. 20 de noviembre II. C.P. 34220, Durango, Durango, México; Phone: (+52) (618) 8-14-20-91 Ext. 82648; FAX: (+52) (618) 8-14-45-40; E-mail: msosam@ipn.mx

0188-4409/$ - see front matter.
http://dx.doi.org/10.1016/j.arcmed.2014.05.007

ethnicity are important risk factors for cardiovascular disease (4,5).

The enzyme CYP1A1 plays an important role in the metabolism of cholesterol and other fatty acids such as arachidonic acid (AA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (6). CYP1A1 acts both as an epoxygenase and ω-1 hydroxylase when converting EPA in 17(*R*), 18(*S*) enantiomers of epoxyeicosatetraenoic acid (17[R]-EETeTr, 18[S]-EETeTr), and 19-OH-EPA metabolites (7,8). CYP1A1 exclusively epoxidizes the ω-3 double bond of EPA and produces 19,20-EDP (9). Furthermore, important contributions of CYP1A1 in the production of signalling and vasoreactive molecules from fatty acids and steroids have been described (10). This suggests that allelic variants of CYP1A1 may affect triglyceride concentrations.

A number of *CYP1A1* allelic variants have been associated with a higher inducibility and/or activity of the enzyme. Among these, *CYP1A1**2C (2455A > G rs1048943, Ile462Val) affects EPA metabolism by altering both, the catalytic efficiency and region specificity of the enzyme (8). Furthermore, CYP1A1*2C has been associated with a 6- to 12-fold higher hydroxylation capacity that forms 17β-estradiol and estrone (11).

Lipid-associated diseases are among the leading causes of morbidity and mortality worldwide. Those conditions have reached populations traditionally considered as healthy such as indigenous groups in Mexico. The ethnic group Tarahumara lives in a mountainous region of Chihuahua in the north of Mexico and represents the second largest Amerindian group in the country (12). Recently, an increase in migration rate to larger settlements because of harsh environmental, social and economic conditions have forced Tarahumaras to adopt a more westernized lifestyle (12,13).

Similarly, the Southern Tepehuano group is also settled in the mountainous region of Durango, Mexico and as happened with Tarahumaras, they have adopted a mestizo lifestyle (14). Thus, this study aimed to investigate the frequencies of *CYP1A1*2C* in Tarahumaras and Tepehuanos and their association with triglyceride levels.

Materials and Methods

Study Subjects and Definitions

We conducted a descriptive and association study including 69 Tarahumara volunteers from ‘‘Choguita’’ community in Chihuahua State and 110 volunteers from the South Tepehuano group with residence in ‘‘Duraznitos’’ community from El Mezquital, Durango and 64 rural Mestizo volunteers from Llano Grande town belonging to the Durango municipality in the state of Durango in the north of Mexico. Moreover, the Amerindian ancestry of the studied groups was confirmed through the analysis of 15 short tandem repeats (STRs) loci (15). This study was approved by the local Ethics Committee of the Hospital General de Durango, Durango, Mexico. All volunteers were thoroughly informed of the study and provided written consent. A questionnaire including dietary habits, alcohol and smoking consumption was applied to all participants. Due to cultural and language barriers, alcoholism was dichotomically evaluated as positive or negative consumption without considering the amount and frequency of consumption; the same criteria were applied for smoking. Family history and a complete medical evaluation were obtained from each volunteer. Each procedure was carried out by the same trained personnel: questionnaire application, medical history, and blood sampling/processing and genotyping.

Biochemical Testing Measurements

Biochemical profile analyses were performed at the Biomedical Research Institute from Universidad Juarez de Durango, Mexico. Normal values are shown in Table 1. Those measurements were done by enzymatic methods (16).

DNA Extraction and Genotyping

Five mL of peripheral venous blood from each volunteer were collected in an EDTA supplemented tube. Genomic DNA was extracted from whole venous blood using a QIAmp DNA Blood Kit according to the manufacturer's instructions (Qiagen, Hilden, Germany). DNA integrity was confirmed by 1% agarose gel electrophoresis and quantified in a NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific, Waltham, MA). Genotyping was done by semi-quantitative Real Time Polymerase Chain Reaction (qRT-PCR) in a StepOne Real Time PCR system (V.2.2, Applied Biosystems, Foster City, CA) under standard conditions. MGB TaqMan probes were used to identify *CYP1A1*2C* (C_25624888_50) polymorphism (17).

Statistical Analysis

Data are presented as mean ± standard deviation or proportions. Median, maximum and minimum values were also

Table 1. Reference values for biochemical testing measurements (16)

Test	Reference values
Creatinine	0.6–1.3 mg/dL
Glucose	70–105 mg/dL
Cholesterol	0–200 mg/dL
Triglycerides	0–150 mg/Dl
AST	0–41 U/L
ALT	0–40 U/L
LDL	35–100 mg/dL
HDL	35–60 mg/Dl
VLDL	0–35 mg/Dl

AST, aspartate aminotransferase; ALT, alanine aminotransferase; LDL, low-density lipoprotein; HDL, high-density lipoprotein; VLDL, very-low-density lipoprotein cholesterol.

calculated. To consider the sample size as valid, we used the Central Limit Theorem in order to avoid bias and support the statistical analyses executed (18−20).

Data were log transformed for normalization of variables. Anthropometric and biochemical data were analyzed using a one-way ANOVA test. Goodness of Fit and Homogeneity analyses as well as Mantel-Haenszel Odds Ratio Estimate were useful to evaluate the association of co-variables with hypertriglyceridemia. Another stratification analysis was done for women of Tepehuano and Tarahumara groups based on the risk age for cardiovascular disease according to different institutions and authors. The cut-off break point for age used in this study was fixed at 45 years old (21). Both allele frequencies and observed and expected heterozygosity were estimated by gene counting method. Hardy-Weinberg Equilibrium (HWE) for genotype distribution was calculated through an exact test using χ^2. Another Pearson χ^2 test was used to estimate differences of the genotype and allele frequencies between ethnic groups. All analyses were done using SPSS Statistic Software v. 17.

AMOVA (Analysis of Molecular Variance) was done in order to explore genetic differences among the populations tested. The Arlequin Software (22) was used for calculation of the AMOVA analysis.

Finally, the association study between genetic and biochemical data was carried out by logistic regression analysis using the free software SNPStats (23). The statistically significant values for all analyses previously mentioned were considered as $p < 0.05$.

Results

In this study, a total of 69 Tarahumaras including 49 women (71%) and 20 men (29%) were analyzed. Their mean age was 44.14 ± 13.02 years old. One hu7ndred ten individuals of Tepehuano origin including 74 women (67%) and 37 men (33%) with an average age of 36.24 ± 13.82 years old were evaluated. Sixty four Mestizo volunteers, 45 women (70%) and 19 men (30%) with an average age of 44.4 ± 14.1 years were also included. Complete profile of anthropometric and biochemical data for the three groups is shown in Table 2. All variables except ALT displayed significant differences between groups ($p < 0.05$).

The analysis by gender revealed significant differences in most of the analyzed variables with the exception of ALT and HDL in men (Table 3). In the case of women, ALT was the only test with no significant differences between groups (Table 3).

In the Tepehuano group, significant differences in number of smokers ($p = 0.000$) and alcohol consumers ($p = 0.000$) between women and men were found. Although Tarahumara showed significant differences in number of smokers according to gender ($p = 0.000$), no significant differences were found in alcohol consumers.

Statistically significant differences between Tarahumara, Tepehuano and Mestizo women were found for waist diameter, BMI, creatinine, cholesterol, triglycerides, AST, LDL HDL and VLDL ($p < 0.05$). Tarahumara and Mestizo women showed the highest values of waist diameter, BMI, triglycerides and VLDL ($p < 0.05$) (Table 3). Mestizo women showed the highest values for age ($p = 0.031$), waist diameter ($p = 0.000$), BMI ($p = 0.000$), cholesterol ($p = 0.000$), triglycerides ($p = 0.03$), LDL ($p = 0.000$) and VLDL ($p = 0.02$) (Table 3). The same comparison in the case of men revealed differences for age ($p = 0.019$), waist circumference ($p = 0.000$), BMI ($p = 0.000$), creatinine ($p = 0.025$), cholesterol ($p = 0.000$), triglycerides ($p = 0.028$), AST ($p = 0.020$), LDL ($p = 0.000$) and VLDL ($p = 0.021$); the remaining variables did not show significant differences between groups.

In order to determine the influence of tobacco and alcohol on triglyceride concentrations, we carried out a further analysis between smokers vs. non-smokers and alcoholics vs. non-alcoholics; this analysis revealed no significant differences in triglycerides or any other of the biochemical parameters evaluated.

Table 2. Biochemical and anthropometric data of Tepehuano, Tarahumara and Mestizo populations

Antropometric-biochemical data	Tepehuano $n = 110$	Tarahumara $n = 69$	Mestizo $n = 64$	p[a]
Age (years)	36.71 ± 13.37	44.14 ± 13.02	44.34 ± 14.13	0.000
Waist (cm)	79.84 ± 8.21	84.85 ± 9.72	94.18 ± 13.30	0.000
Body mass index	21.86 ± 4.44	24.15 ± 4.59	27.93 ± 4.87	0.000
Creatinine (mg/dL)	0.70 ± 0.30	0.79 ± 0.13	0.90 ± 0.19	0.000
Cholesterol (mg/dL)	155.56 ± 40.75	145.63 ± 29.16	203.34 ± 54.30	0.000
Triglycerides (mg/dL)	113.44 ± 66.08	164.12 ± 90.60	177.06 ± 107.64	0.000
AST (u/dL)	35.85 ± 21.40	39.22 ± 12.00	26.81 ± 11.22	0.000
ALT (u/dL)	29.57 ± 31.44	31.76 ± 15.13	30.07 ± 24.69	0.786
LDL (mg/dL)	69.80 ± 22.95	68.59 ± 18.20	118.23 ± 35.30	0.000
HDL (mg/dL)	51.09 ± 12.41	40.99 ± 11.15	50.25 ± 13.62	0.000
VLDL (mg/dL)	22.64 ± 13.17	32.82 ± 18.12	35.68 ± 21.60	0.000

AST, aspartate aminotransferase; ALT, alanine aminotransferase; LDL, low-density lipoprotein; HDL, high-density lipoprotein; VLDL, very-low-density lipoprotein cholesterol.
[a]ANOVA test.

Table 3. Anthropometric and biochemical data of Tepehuano, Tarahumara and Mestizo by gender

	Males				Females			
	Tepehuano = 36	**Tarahumara = 20**	**Mestizo = 19**	***p*[a]**	**Tepehuano = 74**	**Tarahumara = 49**	**Mestizo = 45**	***p*[a]**
Age (years)	36.0 ± 8.68	44.90 ± 13.24	47.53 ± 14.94	0.03	36 ± 14.70	41.26 ± 12.31	43.06 ± 13.75	0.020
Waist (cm)	77.2 ± 8.17	83.51 ± 7.48	91.92 ± 14.09	0.000	79.36 ± 8.66	85.39 ± 9.92	95.11 ± 13.03	0.000
BMI	19.34 ± 5.85	22.44 ± 3.52	26.69 ± 3.97	0.000	22.26 ± 4.39	24.80 ± 4.64	28.44 ± 5.15	0.000
Creatinine (mg/dL)	0.83 ± 0.11	0.82 ± 0.17	0.97 ± 0.27	0.02	0.66 ± 0.35	0.75 ± 0.11	0.88 ± 0.15	0.000
Cholesterol (mg/dL)	147.73 ± 33.34	148.05 ± 33.7	185.84 ± 41.16	0.00	159.07 ± 43.14	144.88 ± 28.36	210.43 ± 57.68	0.000
Triglycerides (mg/dL)	98.6 ± 36.18	135.57 ± 67.62	174.91 ± 123.25	0.03	116.48 ± 72.29	166.72 ± 98.81	177.94 ± 102.11	0.000
AST (U/dL)	38.93 ± 9.35	35.79 ± 14.48	27.45 ± 10.03	0.02	36.83 ± 24.73	38.32 ± 10.82	26.55 ± 11.78	0.003
ALT (U/dL)	29.67 ± 8.87	26.24 ± 10.0	30.81 ± 22.3	0.44	31.52 ± 37.56	32.54 ± 17.09	28.31 ± 18.65	0.750
LDL (mg/dL)	66.13 ± 19.8	69.45 ± 21.58	108.16 ± 24.54	0.000	70.96 ± 24.18	67.74 ± 16.22	122.31 ± 38.30	0.000
HDL (mg/dL)	49.62 ± 11.77	43.30 ± 10.02	45.01 ± 8.5	0.12	53.36 ± 12.91	40.35 ± 10.85	52.37 ± 14.78	0.000
VLDL (mg/dL)	19.72 ± 7.23	26.20 ± 12.61	34.98 ± 24.65	0.02	23.74 ± 14.97	33.34 ± 19.76	35.97 ± 20.50	0.001

Values are mean ± SD.
[a]ANOVA test.

Triglyceride levels in Tarahumara women were independent of age by the Mantel-Haenszel Common Odds Ratio Estimate ($p > 0.05$); conversely Tepehuano females >45 years old have a 3.40 higher chance to present high triglyceride levels than younger Tepehuano women (95% CI, 1.45–7.97).

Genotype and allele frequencies of the three groups are displayed in Table 4. Significant differences were found in *CYP1A1*2C* allele frequency between groups ($p = 0.000$). This polymorphism is in Hardy–Weinberg equilibrium ($p > 0.05$) in the three populations.

After ethnic comparisons, it was found that Tarahumaras have a significant association between high triglyceride levels and *CYP1A1*2C* allele (OR = 2.57; 95% CI 1.12–5.88, $p = 0.024$) under a recessive inheritance model.

In regard to the Tepehuano group, the same analysis showed a significant protective association between normal triglyceride levels and *CYP1A1*2C* polymorphism (OR = 0.28; 95% CI 0.10–0.80, $p = 0.015$) following a dominant inheritance model. In reference to Mestizos, no significant associations were found.

A logistic regression analysis of women of both ethnicities was carried out. After a crude analysis, a strong association between *CYP1A1*2C* and high triglyceride levels was found in Tarahumara women under a recessive inheritance model (OR 4.01, 95% CI 1.39–11.51, $p = 0.007$) (Figure 1). This association remained, after adjustment by age, waist diameter, BMI, tobacco and alcohol consumption, under a recessive inheritance model (OR 7.59, 95% CI 1.93–29.95, $p = 0.0018$).

Discussion

Our results suggest a significant association between *CYP1A1*2C* and high triglyceride levels in Tarahumara women from Chihuahua, Mexico; conversely, a significant

Table 4. Allele and genotype frequencies, heterozygosity (H) and HWE test[a] for *CYP1A1*2C* in Tepehuanos, Tarahumara and Mestizos by gender

CYP1A1	**Males[b]**			**Females[c]**		
Allele	**Tepehuano *n* = 36**	**Tarahumara *n* = 20**	**Mestizo *n* = 19**	**Tepehuano *n* = 74**	**Tarahumara *n* = 49**	**Mestizo *n* = 45**
*1A	38.8 (27.6–50.1)	72.5 (58.7–86.3)	42.1 (26.4–57.8)	35.7 (28.1–43.3)	73.4 (64.7–82.2)	50.0 (39.7–60.3)
*2C	61.1 (49.8–72.4)	27.5 (13.7–41.3)	57.8 (42.2–73.6)	64.3 (56.7–71.8)	26.5 (17.8–35.3)	50.0 (39.7–60.3)
Genotype						
**1 A/*1A*	*n* = 6 He = 15.1; Ho = 11.1	*n* = 10 He = 52.5; Ho = 50.0	*n* = 3 He = 17.7; Ho = 21.0	*n* = 9 He = 12.7; Ho = 12.1	*n* = 26 He = 53.9; Ho = 55.1	*n* = 11 He = 25.0; Ho = 31.1
**1 A/*2C*	*n* = 17 He = 47.5; Ho = 55.5	*n* = 8 He = 39.8; Ho = 45.0	*n* = 10 He = 48.7; Ho = 42.1	*n* = 34 He = 45.9; Ho = 50.0	*n* = 19 He = 38.9; Ho = 36.7	*n* = 23 He = 50.0; Ho = 37.7
**2 C/*2C*	*n* = 13 He = 37.3; Ho = 33.3	*n* = 2 He = 39.8; Ho = 45.0	*n* = 6 He = 33.5; Ho = 36.8	*n* = 31 He = 41.3; Ho = 45.9	*n* = 4 He = 7.0; Ho = 8.1	*n* = 11 He = 25.0; Ho = 31.1

Allele values are % (95% CI), heterozygosity expected (He%), Heterozygosity observed (Ho%) and Hardy-Weinberg equilibrium test.
[a]HWE = $p > 0.05$.
[b]AMOVA test, differences among males $p = 0.000$.
[c]AMOVA test, differences among females $p = 0.000$.

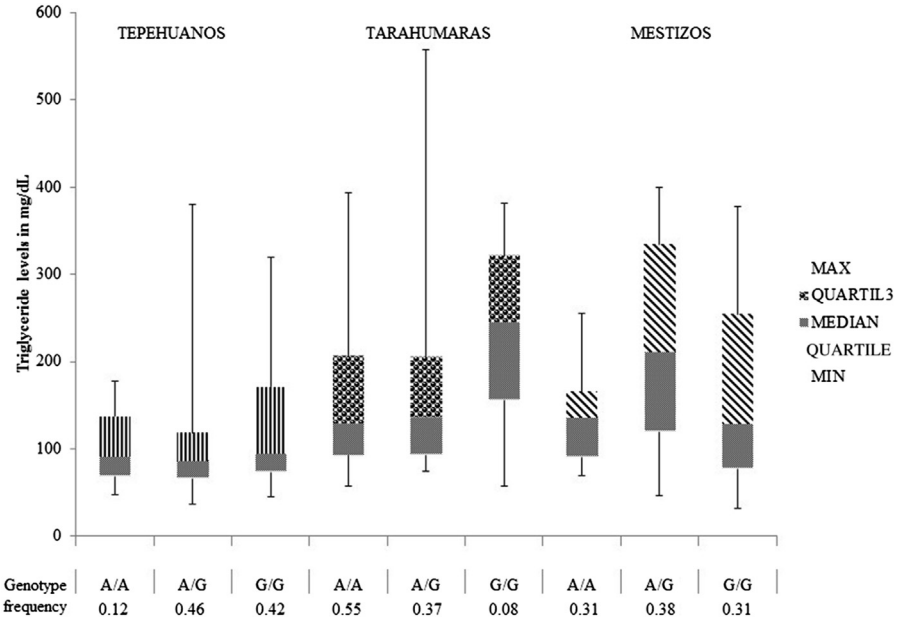

Figure 1. Relationship between triglyceride levels and *CYP1A1*2C* genotypes in Tepehuano, Tarahumara and Mestizo women.

association between *CYP1A1*2C* allele and normal triglyceride levels was observed in Tepehuano women from Durango, Mexico.

Hypertriglyceridemia increases the risk of cardiovascular disease coupled with obesity, metabolic syndrome and type 2 diabetes (24,25). Hypertriglyceridemia affects more than half of the population worldwide (26,27). In Mexico, the incidence of hypertriglyceridemia has been reported to be 31.5% (95% CI, 29.3−33.9) of the total adult population (28,29). Previous reports have evaluated the lipid profile in indigenous populations in Mexico demonstrating an increase in the incidence of hypercholesterolemia and hypertriglyceridemia (14,30,31).

Our results showed significant differences in triglyceride levels between Tarahumaras, Tepehuanos and Mestizos ($p = 0.000$). The observed significant difference between groups was maintained after stratification by gender. The altered lipid profile in Tarahumaras and Tepehuanos may be the result of the acquisition of a westernized lifestyle including processed food and high fat intake with less physical activity (12−14,30−32). However, genetic underlying causes may also play a key role in ailments of lipid metabolism in these ethnic groups.

The frequency of *CYP1A1*2C* in the Asian population (26%) was significantly lower than that observed in Tepehuanos from this study (65%, $p = 0.0005$), but similar to Tarahumaras (27%) (33). The frequency reported in the Inuit population from Canada (45%) is also significantly lower than that of our Tepehuano studied group ($p = 0.0056$) and also different from Tarahumaras ($p = 0.0068$) (34). Remarkably, it has been stated that the Inuit population seems to be protected against cardiovascular disease due to a combination of their traditional dietary lifestyle and genetic traits (35). More importantly, recent findings show that allelic variants like *CYP1A1*2C* involved in the modulation of lipid metabolism may play a crucial role in the normalization of cardiovascular status in the Inuit population (34). Similarly, an evaluation of cardiovascular risk in Taiwanese population revealed a protective dose-response effect of the *CYP1A1*2C* allele (36). Even though this protective effect was also observed in Tepehuanos from Durango, we could not detect it in the Tarahumara population. Lack of a protective effect is probably due to the high triglyceride levels and obesity found in Tarahumara women despite the relatively high frequency of *CYP1A1*2C* polymorphism in this population. Nevertheless, in contrast to the Tarahumaras, the high frequency of *CYP1A1*2C* allele in Mestizos (~50%) was not associated with the high triglyceride levels exhibited by most of this group.

It is noticeable that *CYP1A1*2C* frequency in Tarahumaras was significantly different than that observed in Tepehuanos. This result is in agreement with the differences described between Tarahumaras and other indigenous groups of Mexico in regard to the frequency of polymorphism in *CYP2C9* (37), *CYP2C19* (38) and *CYP2D6* (38,39) genes. This could be a consequence of the genetic differentiation described for the Tarahumara group in addition to their closeness with native North Americans such as NaDene groups (40).

Two potential factors could influence over the observed paradoxical association of *CYP1A1*2C* with hypertriglyceridemia in Tarahumara women: 1) the additive and/or epistatic effect of the *CYP1A1*2C* allele with other genetic polymorphisms, and 2) the small sample size of the

Tarahumara group compared to Tepehuanos. However, the last can be questionable because all the studied populations are in HWE. It is worth mentioning that both indigenous groups exhibited a similar acculturation status, which is related to the increase of high-caloric intake of saturated fat, smoking and alcohol intake (30,41). Therefore, such environmental factors could represent a minor influence in the observed association.

The *CYP1A1* gene and protein expression is upregulated in response to prolonged or chronic exposure to arterial levels of shear stress (hemodynamic biomechanical forces) (42,43); its expression may be important for the maintenance of an athero-protective endothelial phenotype in mouse aortas (42). Thus, the normal triglyceride values in Tepehuanos may be partially explained by this phenomenon.

The role of estrogens in the vascular system is well known by stabilizing endothelial cells and maintaining vascular permeability (44), which maintains an adequate lipoprotein profile. This is lost during menopause and, therefore, the risk of cardiovascular disease increases (41,45). CYP1A1 has a significant role on hydroxylation of 17 β-estradiol (E2) at the C-2, C-4, C-15 α, and C-6 α positions (46). Thus, *CYP1A1*2C* allele may exert a cardioprotective mechanism through modulation of estrogen effects in postmenopausal women (47). In the present study, women of the Tarahumara population presented a marked susceptibility risk for high triglyceride levels associated with the presence of *CYP1A1*2C* polymorphism. An explanation may be that the high triglyceride content in Tarahumara women could saturate the metabolic capacity of the CYP1A1 enzyme. More importantly, the key role of CYP1A1 in estrogen metabolism could explain the observed association in Tarahumara women who are significantly older than the Tepehuano women.

In order to find better explanations of the high lipid profile in Tarahumara women, other genes involved in triglyceride metabolism must be further explored. That is the case of the peroxisome proliferator-activated receptor-γ2 (*PPARγ2*) gene, which may play a central role in the accumulation of abdominal adiposities in Chinese women (48). The combined activity of apolipoprotein C-III (*APOC3*) and apolipoprotein A-I (*APOA1*) genes through C2/S haplotype increase both triglyceride levels and risk of coronary artery disease in Indian (49). Likewise, the apolipoprotein E (*APOE*) gene increases myocardial infarction susceptibility altogether with consumption of saturated fat diets (50). Recently, the fatty acid desaturase 3 (*FADS3*) gene was associated with hyperlipidemia and elevated triglycerides in Mexican populations (51); therefore, it may be advantageous to further study these genes in Amerindian Tarahumaras.

However, some limitations of the study require to be mentioned: a) small sample size in the Tarahumara population as a consequence of the low response rate of these isolated communities, b) lack of quantitative data of alcohol and tobacco consumption due to cultural and language barriers of indigenous individuals at specifying accurate consumption amounts, c) differences in the high frequency of *CYP1A1*2C* allele in Tepehuanos in regard to the lower frequency in Tarahumaras that make necessary an increase in the sample size of the last group, and d) lack of estrogen concentration measurements in females of all the groups evaluated in order to support or discard the hypothesis of the estrogen content as a risk factor for high triglyceride levels.

To our knowledge, this is the first study that reports a strong association between high triglyceride levels and the *CYP1A1*2C* allele in female Tarahumaras from Chihuahua, Mexico. The same allele was significantly associated with normal triglyceride levels in Amerindian Tepehuanos from Durango, Mexico. However, more studies are needed to confirm the genetic role of *CYP1A1* in cardiovascular disease susceptibility.

Acknowledgments

We wish to express our gratitude to all the indigenous volunteers for their collaboration despite the language barrier. We also wish to thank the Molecular Biology Laboratory staff from CIIDIR-DURANGO for all the facilities given, especially to Margarita Ortega, M.S., and the laboratory chief Dr. Carlos Galaviz Hernández. We wish to thank CONACYT-Mexico for the scholarship granted to Claudia Edith Bailón Soto. This study was supported by CONACYT-Mexico Project (2011-C01-162368) and by the SIP IPN Project (20131835). Dr. Martha Sosa-Macías is supported by a grant (COFAA) from the Instituto Politécnico Nacional.

References

1. Austin MA, Hokanson JE, Edwards KL. Hypertriglyceridemia as a cardiovascular risk factor. Am J Cardiol 1998;81:7–12.
2. Pejic RN, Lee DT. Hypertriglyceridemia. J Am Board Fam Med 2006;19:310–316.
3. Bersot T, Haffner S, Harris WS, et al. Hypertriglyceridemia: management of atherogenic dyslipidemia. J Fam Pract 2006;55:1–8.
4. Grundy SM, Cleeman JI, Daniels SR, et al. Diagnosis and Management of the Metabolic Syndrome: An American Heart Association/ National Heart, Lung, and Blood Institute Scientific Statement. Circulation 2005;112:2735–2752.
5. Alberti KG, Zimmet P, Shaw J. Metabolic syndrome—a new worldwide definition. A consensus statement from the International Diabetes Federation. Diabet Med 2006;23:469–480.
6. Arnold C, Konkel A, Fischer R, et al. Cytochrome P450-dependent metabolism of omega-6 and omega-3 long-chain polyunsaturated fatty acids. Pharmacol Rep 2010;62:536–547.
7. Schwarz D, Kisselev P, Ericksen SS, et al. Arachidonic and eicosapentaenoic acid metabolism by human CYP1A1: highly stereoselective formation of 17(R),18(S)-epoxyeicosatetraenoic acid. Biochem Pharmacol 2004;67:1445–1457.
8. Schwarz D, Kisselev P, Chernogolov A, et al. Human CYP1A1 variants lead to differential eicosapentaenoic acid metabolite patterns. Biochem Biophys Res Commun 2005;336:779–783.
9. Fer M, Dréano Y, Lucas D, et al. Metabolism of eicosapentaenoic and docosahexaenoic acids by recombinant human cytochromes P450. Arch Biochem Biophys 2008;471:116–125.

10. Choudhary D, Jansson I, Stoilov I, et al. Metabolism of retinoids and arachidonic acid by human and mouse cytochrome P450 1b1. Drug Metab Dispos 2004;32:840−847.
11. Kisselev P, Schunck WH, Roots I, et al. Association of CYP1A1 polymorphisms with differential metabolic activation of 17beta-estradiol and estrone. Cancer Res 2005;65:2972−2978.
12. Comisión Nacional para el Desarrollo de los Pueblos Indígenas. CDI. 2013. Tarahumaras-rarámuri and Tepehuanos del Sur O'dam [online]. Available from: http://www.cdi.gob.mx/index.php?option = com_content&task = view&id = 609&Itemid = 62 [Accessed 2 May 2013].
13. Pérez-Escamilla R, Álvarez Uribe MC, Segall-Correa AM, et al. Escala Latinoamericana y Caribeña de Seguridad Alimentaria (ELCSA). Memorias de la 1a Conferencia en América Latina y el Caribe sobre la medición de la seguridad alimentaria en el hogar. Perspectivas en Nutrición Humana 2007;(S):117−134.
14. Rodríguez-Morán M, Guerrero-Romero F, Brito-Zurita O, et al. Cardiovascular risk factors and acculturation in Yaquis and Tepehuanos Indians from Mexico. Arch Med Res 2008;39:352−357.
15. Rangel-Villalobos H, Martínez-Sevilla VM, Salazar-Flores J, et al. Forensic parameters for 15 STRs in eight Amerindian populations from the north and west of Mexico. Forensic Sci Int Genet 2013;3:e62−e65.
16. Ervin RB. Prevalence of Metabolic Syndrome Among Adults 20 Years of Age and Over, by Sex, Age, Race and Ethnicity, and Body Mass Index: United States, 2003−2006. Division of Health and Nutrition Examination Surveys. NHANES Survey. Number 15. May 5, 2009 [online]. Available from: http://www.cdc.gov/nchs/nhanes.htm (NHANES) 2003−2006. [Accessed August 4, 2013].
17. Applied Biosystems protocols 2010. Applied Biosystems StepOne v2.2. Genotyping Experiments. Part Number 4376786 Rev. F
18. Alvarado H, Batanero C. Significado del Teorema del Límite Central en textos universitarios de Probabilidad y Estadística. Estud. pedagóg [online]. 2008, vol. 34, n.2, pp. 7−28. ISSN 0718-0705.
19. Devore J. Probabilidad y Estadística para Ingeniería y Ciencias. 5th ed México: Thompson; 2001.
20. Johnson R, Kuby P, eds. Estadística Elemental. 3rd ed. México: Thompson; 2004.
21. Escobedo-de la Peña J, Rodríguez-Ábrego G, Buitrón-Granados LV. Morbilidad y mortalidad por cardiopatía isquémica en el Instituto Mexicano del Seguro Social. Estudio ecológico de tendencias en población amparada por el Instituto Mexicano del Seguro Social entre 1990 y 2008. Arch Cardiol Mex 2010;80:242−248.
22. Excoffier L, Laval G, Schneider S. Arlequin ver. 3.0: An integrated software package for population genetics data analysis. Evol Bioinform Online 2005;1:47−50.
23. Solé X, Guinó E, Valls J, et al. SNPStats: a web tool for the analysis of association studies. Bioinformatics 2006;22:1928−1929. http://dx.doi.org/10.1093/bioinformatics/btl268. [online]. Available from: http://bioinfo.iconcologia.net/SNPStats. [Accessed 2012, 2013 and 2014].
24. Lakshmi SV, Naushad SM, Saumya K, et al. Role of CYP1A1 haplotypes in modulating susceptibility to coronary artery disease. Indian J Biochem Biophys 2012;5:349−355.
25. Hodis HN, Mack WJ, Krauss RM, et al. Pathophysiology of triglyceride-rich lipoproteins in atherothrombosis: clinical aspects. Clin Cardiol 1999;22:15−20.
26. Lozano R, Murray CJL, Lopez AD, et al. Miscoding and misclassification of ischemic heart disease mortality. Global Programme on Evidence for Health Policy Working Paper No. 12. World Health Organization; 2001. [online] Available from: http://www.who.int/healthinfo/paper12.pdf. [Accessed 11 September 2013].
27. WHO 2013. World Health Organization. A global brief on hypertension. Silent killer, global public health crisis. [online] Available from: http://apps.who.int/iris/bitstream/10665/79059/1/WHO_DCO_WHD_2013.2_eng.pdf [Accessed September 11 2013].
28. ENSANUT 2012. Encuesta Nacional de Salud y Nutrición 2012 [online]. Available from: http://ensanut.insp.mx/doctos/ENSANUT2012_PresentacionOficialCorta_09Nov2012.pdf. [Accessed 11 July 2013].
29. Aguilar-Salinas CA, Gómez-Pérez FJ, Rull J, et al. Prevalence of dyslipidemias in the Mexican National Health and Nutrition Survey 2006. Salud Publica Mex 2006;52:44−53.
30. Leal-Berumen I, Santana-Rodriguez V, Hernández-Rodríguez P, et al. Screening for metabolic syndrome risk factors in mestizo, tarahumara and mennonite scholars from Chihuahua Mexico. BMC Proc 2012;6:31.
31. McMurry MP, Cerqueira MT, Connor SL, et al. Changes in lipid and lipoprotein levels and body weight in Tarahumara Indians after consumption of an affluent diet. N Engl J Med 1991;325:1704−1708.
32. McMurry MP, Connor WE, Cerqueira MT. Dietary cholesterol and the plasma lipids and lipoproteins in the Tarahumara Indians: a people habituated to a low cholesterol diet after weaning. Am J Clin Nutr 1982;35:741−744.
33. Cosma G, Crofts F, Taioli E, et al. Relationship between genotype and function of the human CYP1A1 gene. J Toxicol Environ Health 1993;40:309−316.
34. Rudkowska I, Dewailly E, Hegele RA, et al. Gene−diet interactions on plasma lipid levels in the Inuit population. Br J Nutr 2013;109:953−961.
35. Counil E, Julien P, Lamarche B, et al. Association between trans-fatty acids in erythrocytes and pro-atherogenic lipid profiles among Canadian Inuit of Nunavik: possible influences of sex and age. Br J Nutr 2009;102:766−776.
36. Yeh CC, Sung FC, Kuo LT, et al. Polymorphisms of cytochrome P450 1A1, cigarette smoking and risk of coronary artery disease. Mutat Res 2009;667:77−81.
37. Sosa-Macías M, Lazalde-Ramos BP, Galaviz-Hernández C, et al. Influence of admixture components on CYP2C9*2 allele frequency in eight indigenous populations from Northwest Mexico. Pharmacogenomics J 2013;13:567−572.
38. Salazar-Flores J, Torres-Reyes LA, Martínez-Cortés G, et al. Distribution of CYP2D6 and CYP2C19 polymorphisms associated with poor metabolizer phenotype in five Amerindian groups and western Mestizos from Mexico. Genet Test Mol Biomarkers 2012;16:1098−1104.
39. Lazalde-Ramos BP, Martínez-Fierro Mde L, Galaviz-Hernández C, et al. CYP2D6 gene polymorphisms and predicted phenotypes in eight indigenous groups from northwestern Mexico. Pharmacogenomics 2014;15:339−348.
40. Rangel-Villalobos H, Muñoz-Valle JF, González-Martín A, et al. Genetic admixture, relatedness, and structure patterns among Mexican populations revealed by the Y-chromosome. Am J Phys Anthropol 2008;135:448−461.
41. Rodríquez-Morán M, Guerrero-Romero F, Rascón-Pacheco RA, et al. Dietary factors related to the increase of cardiovascular risk factors in traditional Tepehuanos communities from Mexico. A 10 year follow-up study. Nutr Metab Cardiovasc Dis 2009;19:409−416.
42. Conway DE, Sakurai Y, Weiss D, et al. Expression of CYP1A1 and CYP1B1 in human endothelial cells: regulation by fluid shear stress. Cardiovasc Res 2009;81:669−677.
43. Han Z, Miwa Y, Obikane H, et al. Aryl hydrocarbon receptor mediates laminar fluid shear stress-induced CYP1A1 activation and cell cycle arrest in vascular endothelial cells. Cardiovasc Res 2008;77:809−818.
44. Darkow DJ, Lu L, White RE. Estrogen relaxation of coronary smooth muscle is mediated by nitric oxide and cGMP. Am J Physiol 1997;272:2765−2773.

45. Reddy KS, Chandala SR. A comparative study of lipid profile and oestradiol in pre- and post-menopausal Women. J Clin Diagn Res 2013;7:1596−1598.
46. Spink DC, Eugster HP, Lincoln DW 2nd, et al. 17β-Estradiol hydroxylation catalyzed by human cytochrome P450 1A1: a comparison of the activities induced by 2,3,7,8-tetrachlorodibenzo-p-dioxin in MCF-7 cells with those from heterologous expression of the cDNA. Arch Biochem Biophys 1992;293:342−348.
47. Almeida S, Zandoná MR, Franken N, et al. Estrogen-metabolizing gene polymorphisms and lipid levels in women with different hormonal status. Pharmacogenomics J 2005;5:346−351.
48. Li H, Chen X, Guan L, et al. MiRNA-181a regulates adipogenesis by targeting tumor necrosis factor-α (TNF-α) in the porcine model. PLoS One 2013;8:e71568.
49. AshokKumar M, Subhashini NG, SaiBabu R, et al. Genetic variants on apolipoprotein gene cluster influence triglycerides with a risk of coronary artery disease among Indians. Mol Biol Rep 2010;37:521−527.
50. Yuan G, Al-Shali ZK, Hegele RA. Hypertriglyceridemia: its etiology, effects and treatment. CMAJ 2007;176:1113−1120.
51. Plaisier CL, Horvath S, Huertas-Vazquez A, et al. A systems genetics approach implicates USF1, FADS3, and other causal candidate genes for familial combined hyperlipidemia. PLoS Genet 2009;5:e1000642.