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ALLELE AND GENOTYPE FREQUENCIES OF CYP2B6 AND CYP2C19 POLYMORPHISMS IN EGYPTIAN AGRICULTURAL WORKERS

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Abstract

Genetic variability in cytochrome P-450 (CYP) has the potential to modify pharmacological and toxicological responses to many chemicals. Both CYP2B6 and CYP2C19 are pharmacologically and toxicologically relevant due to their ability to metabolize multiple drugs and environmental contaminants, including the organophosphorus (OP) pesticide chlorpyrifos. The aim of this study was to determine the prevalence of CYP2B6 and CYP2C19 variants in an indigenous Egyptian population ($n = 120$) that was shown to be occupationally exposed to chlorpyrifos. Further, the genotyping data was compared for Egyptians with previously studied populations to determine between population differences. Allelic frequencies were CYP2B6 1459C > T (3.8%), CYP2B6 785A > G (30.4%), CYP2B6 516G > T (28.8%), CYP2C19 681G > A (3.8%), and CYP2C19 431G > A (0%). The most prevalent CYP2B6 genotype combinations were CYP2B6 *1/*1 (44%), *1/*6 (38%), *6/*6 (8%), and *1/*5 (6%). The frequency of the CYP2C19 genotype combinations were CYP2C19 *1/*1 (93%), *1/*2 (6%), and *2/*2 (1%). The frequency of the CYP2B6 516G > T and CYP2B6 785A > G polymorphisms in this Egyptian cohort is similar to that found North American and European populations but significantly different from that reported for West African populations, while that of CYP2B6 1459C > T is similar to that found in Africans and African Americans. The observed frequency of CYP2C19 681G > A in Egyptians is similar to that of African pygmies but significantly different from other world populations, while CYP2C19 431G > A was significantly different from that of African pygmies but similar to other world populations.

Characterization of pharmacologically relevant polymorphisms in cytochrome P-450s (CYPs) has great potential to improve drug efficacy and reduce toxicity (Bozina et al. 2009; Zanger et al. 2007). Similarly, genetic variability in CYP has the potential to modify the bio-

effective dose and resulting toxicity from environmental and occupational chemical exposures. CYP2B6 and CYP2C19 are two pharmacologically and toxicologically relevant CYPs due to their ability to metabolize several different drugs/toxicant and their highly polymorphic nature (Bozina et al. 2009; Zanger et al. 2007). CYP2B6 metabolizes drugs such as the non-nucleoside reverse transcriptase inhibitor efavirenz (Ward et al. 2003), the cytostatic prodrug cyclophosphamide (Chang et al. 1993), and the antimalarial artemisinin (Svensson et al. 1999). CYP2C19 metabolizes a variety of drugs such as the anti-ulcer drug omeprazole (Andersson et al. 1992), the anticonvulsant mephenytoin (Daniel et al. 1996), and the anxiolytic drug diazepam (Galetin et al. 2006). Both CYP2B6 and CYP2C19 are also active in the metabolism of a variety of environmental contaminants (Povey 2010), including the organophosphorus (OP) pesticide chlorpyrifos (Foxenberg et al. 2007). Chlorpyrifos may be activated to a potent β -esterase inhibitor by CYP2B6 or detoxified to a dearylated metabolite by CYP2C19 (Foxenberg et al. 2007). The relative balance between the formation of activated and detoxified metabolites of chlorpyrifos is potentially influenced by age, gender, CYP profiles, and CYP polymorphisms (Atterberry et al. 1997; Ma et al. 1994; Rose et al. 2005).

Several single-nucleotide polymorphisms (SNP) were identified in the CYP2B6 and CYP2C19 genes. Lang et al. (2001) first identified nine SNP in the CYP2B6 gene of German Caucasians, of which five result in amino acid substitutions. Several of the identified SNP are common to multiple CYP alleles and haplotypes (Lang et al. 2001). The CYP2B6*5 (1459C > T) genotype is associated with decreased activity and protein (Lang et al. 2001), while the CYP2B6*4 (785A > G) genotype is associated with increased CYP2B6 activity (Jinno et al. 2003). The CYP2B6*6 (516G > T and 785A > G) genotype has been associated with decreased protein expression (Desta et al. 2007) and decreased activity toward *S*-mephenytoin (Lang et al. 2001) and efavirenz (Desta et al. 2007) but increased activity toward 7-ethoxy-4-trifluoro-methylcoumarin (Jinno et al. 2003) and cyclophosphamide (Xie et al. 2003), suggesting that the functional change is substrate specific. The CYP2C19 variant CYP2C19*2 (681G > A) leads to an aberrant splice site resulting in a null CYP2C19 allele (Demorais et al. 1994). The CYP2C19*9 (431G > A) allele results in an amino acid change that is associated with decreased CYP2C19 activity (Blaisdell et al. 2002).

The aim of the present study was to determine the frequency of CYP2B6 (516G > T, 785A > T and 1459C > T) and CYP2C19 (681G > A and 431G > A) polymorphisms within a cohort of male Egyptian pesticide applicators with documented occupational exposures to chlorpyrifos (Farahat et al. 2010; 2011). This is the first time that these CYP2B6 and CYP2C19 polymorphisms have been studied in an Egyptian population, with the exception of one study that determined the frequency of CYP2C19 681 G > A (Hamdy et al. 2002).

METHODS

Study Setting and Population

The study setting was previously described (Farahat et al. 2010). In brief, the study took place in one of 29 governorates in Egypt, Menoufia, which is situated in the Nile River delta north of Cairo. Male agricultural workers aged 14 to 69 yr were asked to donate saliva during enrollment of the study. In total, 120 participants (96% being agricultural workers) donated saliva at enrollment and were similar in terms of body mass index (BMI), ethnicity, and gender (data not shown). All participants were Egyptian, indigenous to the Nile delta region, with the vast majority being born, raised, and currently residing in Menoufia, Egypt. The protocol and consent forms used in this research were approved by the Oregon Health & Science University (USA) and Menoufia University (Egypt) institutional review boards. Informed consent was obtained from each worker before the start of the study.

Saliva Collection and DNA Isolation

Approximately 1-2 ml of saliva for each subject was collected using an OG-250 saliva collection kit (DNA Genotek Inc., Kanata, ON). Collection kits containing saliva samples were transported to Menoufia University (Shebin El-Kom, Egypt), where they were stored at room temperature prior to analysis. Genomic DNA was isolated from saliva following the manufacturer's instructions (DNA Genotek Inc., Kanata, ON). DNA concentration was determined using a nanodrop spectrophotometer (ThermoFisher Scientific, Waltham, MA), and only quality DNA (A260/280 ratio > 1.7) was used for genotyping analysis. Purified DNA was stored in TE buffer (10 mM Tris-HCl, 1 mM ethylene-diamine tetraacetic acid [EDTA], pH 8.0) at -20°C.

Multiplex Amplification of CYP2B6, CYP2C19, and β -Actin Exon Fragments

CYP2B6 (exon 5 + 6, exon 9, and exon 4), CYP2C19 (exon 2 + 3 and exon 5), and β -actin (exon 4) DNA fragments were amplified by multiplex polymerase chain reaction (PCR) (Griese et al. 1999; Jacob et al. 2004), using PCR primers previously described for CYP2B6 (Jacob et al. 2004) and CYP2C19 (Blaisdell et al. 2002). The primer sequence for β -actin was ATGTCCCCCGTCTGGCCTGG (forward) and CTGTAGCCGCGCTCGGTGAG (reverse). The multiplex PCR reaction (25 μ l total volume) contained approximately 20–500 ng genomic DNA, 12.5 μ l Qiagen Multiplex PCR buffer (Qiagen Inc., Valencia, CA), 0.2 μ M of each primer, and 9 μ l PCR-grade water. The multiplex PCR reaction was performed in a Techne TC-412 thermocycler (Barloworld Scientific Ltd., Staffordshire, UK) using the following thermocycle conditions: pre-denaturation at 95°C for 15 min, 35 cycles of amplification (94°C for 30 s, 61°C for 1 min 30 s, 72°C for 1 min 45 s), and a final extension at 72°C for 10 min. To verify the presence of each amplicon, an aliquot of the multiplex PCR products was separated by electrophoresis on a 2% agarose gel containing ethidium bromide and visualized on an ultraviolet (UV) transilluminator.

Allele-Specific Multiplex PCR Genotyping

Allele-specific PCR assays were conducted for the following CYP polymorphisms: CYP2B6 516G > T, CYP2B6 785A > G, CYP2B6 1459C > T, CYP2C19 431G > A, and CYP2C19 681G > A. In addition, β -actin was amplified in all reactions as an internal positive control. For each individual to be genotyped, two different multiplex PCR reactions were conducted, which contained either PCR primers for the dominant CYP alleles or the variant CYP alleles. The allele-specific PCR primers have previously been described for CYP2B6 SNPs (Jacob et al. 2004) and the CYP2C19 681 G > A SNP (Griese et al. 1999). The sequences for the remaining allele-specific PCR primers were: CYP2C19 431G > A, CAGCGGGCTTCCTCTTGAACCC (G allele), A GCGGGCTTCCTCTTGAACCT (A allele), ATC CGGCGTTTCTCCCTCAT (consensus); β -actin, GGGGAGGGCATACCCCTCGT (forward), TGTCCCCAGTGGCTTCCCCA (reverse). The multiplex allele-specific PCR reaction (25 μ l total volume) contained 1 μ l of a 1:100 dilution of PCR product from the first round of multiplex PCR (i.e., CYP exon amplification product diluted in PCR-grade water), 12.5 μ l Qiagen Multiplex PCR buffer (Qiagen, Inc., Valencia, CA), 0.2 μ M of each primer, and 9 μ l PCR-grade water. Thermocycling conditions were: pre-denaturation at 95°C for 15 min, 18 cycles of amplification (94°C for 30 s, 59°C for 30 s, 72°C for 30 s), and a final extension at 72°C for 10 min. An aliquot of PCR product from the wild-type and one from the variant PCR reactions were separated side-by-side by electrophoresis on a 3% agarose gel containing ethidium bromide and visualized on a UV transilluminator. Genotypes were then inferred by comparing the banding pattern for wild-type and variant PCR reactions for each individual's sample.

Quality control measures included: (1) direct DNA sequencing of a subset of samples to confirm the inferred genotype detected by the allele-specific PCR assay; (2) repeated PCR-

based genotyping of select DNA samples to test for concordance in genotype; and (3) repeated analysis for all low-confidence samples until a reliable genotype could be determined.

Statistical Analysis

Allele and genotype frequencies for the SNPs investigated were determined using the SNP and disease association analysis software SNPalyze (version 7, Dynacom Co. Ltd., Yokohama, Japan). Deviations from Hardy–Weinberg equilibrium expectations were determined using the chi-squared test. Allele frequencies from the current study were compared with published data using a chi-squared test. A *p* value .05 was considered significant unless otherwise noted.

RESULTS

The CYP2B6 SNP 516G > T, 785A > G, and 1459C > T and the CYP2C19 SNP 431G > A and 681G > A were investigated in this study. All of these polymorphisms, with the exception of the variant CYP2C19 431G > A, were observed in this Egyptian population (Table 1) and were consistent with Hardy–Weinberg equilibrium. For all of the CYP2B6 and CYP2C19 genotypes, the variant alleles (1459T, 785G, 516T, 681A, 431A) occurred with a lower frequency than the wild-type alleles. CYP2B6 785A > G and CYP2B6 516G > T showed the greatest variation within the population. The frequencies of the CYP2B6 785 alleles were A = 0.70, G = 0.30, while the frequencies of the CYP2B6 516 alleles were G = 0.71, T = 0.29. A relatively large frequency of the population was heterozygous for CYP2B6 785A > G (0.41) and CYP2B6 516G > T (0.41), while the heterozygous frequencies were small for CYP2B6 1459C > T (0.08), CYP2C19 681G > A (0.06), and CYP2C19 431G > A (0).

For CYP2B6 the *1 (wild-type), *4 (785A > G), *5 (1459C > T), and *6 (516G > T and 785A > G) genotypes were identified in our study population. The most frequent CYP2B6 haplotype combinations were *1/*1 (44%), *1/*6 (38%), *6/*6 (8%), and *1/*5 (6%) (Table 2). The 6 other CYP2B6 haplotype combinations identified in this cohort occurred in less than 2% of the population. For CYP2C19, the *1 (wild-type) and *2 (681G > A) genotypes were identified with haplotype combinations of *1/*1 (93%), *1/*2 (6%), and *2/*2 (0.8%) (Table 2). The percentages for the combined CYP2C19 and CYP2B6 genotype for the eight individuals who contained a CYP2C19*2 genotype were CYP2C19*2/CYP2B6*1 (38%), CYP2C19*2/CYP2B6*4 (13%), CYP2C19*2/CYP2B6*5 (0%) and CYP2C19*2/CYP2B6*6 (50%).

DISCUSSION

The present study determined the genotype distribution of CYP2B6 and CYP2C19, two pharmacologically and toxicologically relevant genes, among an Egyptian agricultural population. This Egyptian population was chosen for two primary reasons, with the first being that there is currently a lack of genotyping data for Egyptians and the second that this population is potentially exposed to environmental contaminants such as OP pesticides (Farahat et al. 2010; 2011), which are metabolized by CYP2B6 and CYP2C19 (Ellison et al. 2011; Foxenberg et al. 2007). The availability of genotyping data for this population will provide novel insight into the ethnic diversity of Egyptians.

The observed frequencies of CYP2B6 516G > T and CYP2B6 785A > G in Egyptians was 0.29 and 0.3, respectively, which is similar to North American and European populations but significantly different from West African populations (Table 3). The frequency of CYP2B6 1459C > T in Egyptians was 0.04, which is comparable to Africans, African-Americans, and

Asians (Table 3). Polymorphisms in CYP2B6 might alter the pharmacokinetics of several drugs, such as efavirenz and cyclophosphamide, both of which are metabolized by CYP2B6 (Chang et al. 1993; Ward et al. 2003). Efavirenz is a non-nucleoside reverse transcriptase inhibitor often used as part of a combination therapy for HIV patients. Efavirenz has a narrow therapeutic window, with elevated plasma concentrations being associated with central nervous system (CNS)-related side effects such as dizziness, depression, and difficulty sleeping (Marzolini et al. 2001). Several studies consistently found that the CYP2B6 515G > T polymorphism is associated with higher plasma concentrations and the appearance of CNS-related side effects (Haas et al. 2004; Ribaudo et al. 2006). Cyclophosphamide is a widely used anticancer and immunosuppressant pro-drug that requires metabolic activation, primarily by CYP2B6, to form its active metabolite (Chang et al. 1993). The CYP2B6 515G > T polymorphism is associated with increased cyclophosphamide activity and clearance compared to the wild-type gene (Nakajima et al. 2007; Xie et al. 2006). Ariyoshi et al. (2011) demonstrated that the CYP2B6 785A > G polymorphism results in increased metabolic activity toward efavirenz and decreased activity toward cyclophosphamide; however, when both the CYP2B6 785A > G and CYP2B6 516G > T polymorphisms are present the metabolic profile is the opposite. The effects of the CYP2B6 516G > T polymorphism on efavirenz and cyclophosphamide metabolism and efficacy appear to be in contrast to each other, suggesting that the functional change of the CYP2B6 516G > T polymorphism is substrate specific. Furthermore, the CYP2B6 516G > T polymorphism can inverse the effects produced by the CYP2B6 785A > G polymorphisms for certain drugs (Ariyoshi et al. 2011).

The observed frequency of CYP2C19 681G > A within the Egyptian study population was 0.04, which is similar to that previously reported for a Ghanaian population (Kudzi et al. 2009); however, it is significantly lower than that reported for other African, Asian, and European populations (Table 4). The difference in the frequency of CYP2C19 681G > A among Egyptians and other African populations highlights the importance of studying gene polymorphisms on a population specific basis. Few studies examined the prevalence of the CYP2C19 431G > A polymorphism, which is associated with the CYP2C19*9 genotype (Blaisdell et al. 2002). This SNP was found to be present in 17% of African pygmies but absent in African-Americans, Asians, and Caucasians (Blaisdell et al. 2002). CYP2C19 431G > A variant was not detected within our study population, but this needs to be confirmed in a larger Egyptian population. More studies need to be conducted to determine the frequency of this SNP among different populations globally. CYP2C19 is clinically important for the metabolism of drugs such as the anticonvulsant mephenytoin and the anti-ulcer drug omeprazole (Andersson et al. 1992; Daniel et al. 1996). Polymorphisms in CYP2C19 may result in a poor metabolizer phenotype for both mephenytoin and omeprazole, as well as other drugs metabolized by CYP2C19.

Organophosphorus (OP) pesticides, such as chlorpyrifos, are known to be metabolized by CYP, with CYP2B6 and CYP2C19 being the main enzymes involved in OP pesticide activation and detoxification, respectively (Foxenberg et al. 2007). Currently few data are available on the impact of CYP2B6 and CYP2C19 polymorphisms on the in vitro metabolism of chlorpyrifos or other OP pesticides. Human liver microsomes genotyped as CYP2B6*6 (516G > T and 785A > G) display a nonsignificant increase in the formation of the biologically active metabolite of chlorpyrifos compared to microsomes with a wildtype genotype (Croom et al. 2010). Few investigations examined the in vivo influence of CYP polymorphisms on OP pesticide toxicity. Povey et al. (2007) examined the ill health of farmers exposed to OP pesticide-containing sheep dip and saw numerically elevated risks associated with the CYP2D6 wildtype genotype. The study did not examine other CYP isoforms as potential genetic risk factors, so it is unclear how variants of CYP2B6 and CYP2C19 would correlate with worker health. Foxenberg et al. (2011) utilized a

physiologically based pharmacokinetic and pharmacodynamic model to demonstrate that variation in CYP2B6 and CYP2C19 hepatic content might influence OP pesticide related toxicity. Further study needs be conducted to determine the effects of CYP polymorphisms on OP pesticide metabolism and toxicity. It is possible that interindividual differences in susceptibility to OP pesticide toxicity may in part be mediated by the CYP genotype and phenotype of an individual.

In conclusion, the prevalence of pharmacologically and toxicologically relevant CYP2B6 and CYP2C19 polymorphisms was determined in a population of indigenous Egyptians. The frequency of CYP2B6 516G > T and CYP2B6 786A > G in Egyptians was similar to that reported for North American and European populations (Haas et al. 2004; Jacob et al. 2004) but significantly different from West African populations (Mehlotra et al. 2006), while the CYP2B6 1459C > T SNP was similar to Africans and African-Americans (Haas et al. 2004; Klein et al. 2005; Mehlotra et al. 2006). The observed frequency of CYP2C19 681G > A in Egyptians was similar to African pygmies but significantly different from most other world populations, while CYP2C19 431G > A was significantly different from African pygmies but similar to other world populations (Blaisdell et al. 2002; Goldstein et al. 1997; Kudzi et al. 2009). These results provide novel information on the prevalence of polymorphisms in key enzymes involved in OP pesticide metabolism within a population that may have neurobehavioral deficits as a consequence of occupational exposure to OP pesticides (Abdel Rasoul et al. 2008; Clegg et al. 1999; Cole et al. 1998; Farahat et al. 2003; 2011). CYP2B6 and CYP2C19 genotypes may serve as a useful biomarker of genetic susceptibility toward OP pesticide toxicity; however, further investigation is needed to validate their usefulness in this context.

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TABLE 1

Allele and Genotype Frequencies for CYP2B6 and CYP2C19 Polymorphisms in the Egyptian Study Population

	Number of individuals (total = 120)	Frequency (%)	95% CI
CYP2B6 1459 C > T(rs3211371)			
Genotype			
WT/WT	111	92.5	87.8-97.2
WT/VT	9	7.50	2.79-12.2
VT/VT	0	0	–
Allele			
C		96.3	93.5-98.5
T		3.80	1.52-6.48
CYP2B6 785 A > G (rs2279343)			
Genotype			
WT/WT	59	49.2	40.2-59.1
WT/VT	49	40.8	32.0-49.6
VT/VT	12	10.0	4.63-15.4
Allele			
A		69.6	64.2 -75.8
G		30.4	24.2 -35.8
CYP2B6 516 G > T (rs3745274)			
Genotype			
WT/WT	61	50.8	41.9-59.8
WT/VT	49	40.8	32.0-49.6
VT/VT	10	8.30	3.39-13.3
Allele			
G		71.3	65.3 -76.7
T		28.8	23.3 -34.7
CYP2C19 681 G > A (rs4244285)			
Genotype			
WT/WT	112	93.3	88.9 -97.8
WT/VT	7	5.80	1.64-10.0
VT/VT	1	0.80	0 - 2.45
Allele			
G		96.3	93.5 -98.5
A		3.80	1.52-6.48
CYP2C19 431 G > A (rs17884712)			
Genotype			
WT/WT	120	100	–
WT/VT	0	0	–
VT/VT	0	0	–

	Number of individuals (total = 120)	Frequency (%)	95% CI
Allele			
G		100	–
A		0	–

Note. CI, confidence interval; WT, wild type; VT, variant.

TABLE 2

CYP2B6 and CYP2C19 Genotype Combinations Within the Egyptian Study Population

Genotype combination	Number of individuals (total = 120)	Frequency (%)
CYP2B6		
*1/*1	53	44.2
*1/*4	1	0.83
*1/*5	7	5.83
*1/*6	45	37.5
*4/*4	1	0.83
*4/*5	0	0
*4/*6	2	1.67
*5/*5	0	0
*5/*6	2	1.67
*6/*6	9	7.5
CYP2C19		
*1/*1	112	93.3
*1/*2	7	5.8
*2/*2	1	0.8

TABLE 3

Allele Frequencies of CYP2B6 Polymorphisms Among the Egyptian Study Population and Other Previously Studied Populations

Population	516 G > T		785 A > G		1459 C > T		Reference
	G	T	A	G	C	T	
Africa							
Egyptian (<i>n</i> = 120)	0.71	0.29	0.70	0.30	0.96	0.04	Current study
Ghanaian (<i>n</i> = 82)	0.51 ^a	0.49 ^a	0.52 ^a	0.48 ^a	0.97	0.03	(Klein et al. 2005)
West African (<i>n</i> = 166)	0.58 ^a	0.42 ^a	0.58 ^a	0.42 ^a	0.98	0.02	(Mehloua et al. 2006)
North America							
African American (<i>n</i> = 50)	0.62	0.38	–	–	0.99	0.01	(Haas etal. 2004)
European American (<i>n</i> = 89)	0.78	0.22	–	–	0.88	0.12	(Haas etal. 2004)
Europe							
British (<i>n</i> = 135)	0.72	0.28	0.70	0.30	0.88 ^a	0.12 ^a	(Jacob etal. 2004)
German Caucasian (<i>n</i> = 215)	0.71	0.29	0.67	0.33	0.86 ^a	0.14 ^a	(Lang etal. 2001)
Swiss (<i>n</i> = 209)	0.76	0.24	0.74	0.26	0.89 ^a	0.11 ^a	(Crettol etal. 2005)
Asia							
Korean (<i>n</i> = 316)	0.86 ^b	0.14 ^b	0.81 ^a	0.19 ^a	0.99	0.01	(Cho et al. 2004)
Chinese (<i>n</i> = 386)	0.79	0.21	0.72	0.28	0.997	0.003	(Guan etal. 2006)

^aSignificant at *p* < .05 when compared with Egyptians.

^bSignificant at *p* < .001 when compared with Egyptians.

TABLE 4
Allele Frequencies of CYP2C19 Polymorphisms Among the Egyptian Study Population and Other Previously Studied Populations

Population	681 G > A		431 G > A		Reference
	G	A	G	A	
Africa					
Egyptian (<i>n</i> = 120)	0.96	0.04	1.00	0	Current study
Ghanaian (<i>n</i> = 204)	0.94	0.06	–	–	(Kudzi et al. 2009)
African Pygmies (<i>n</i> = 9)	1	0	0.83 ^b	0.17 ^b	(Blaisdell et al. 2002)
Ethiopian (<i>n</i> = 110)	0.87 ^a	0.13 ^a	–	–	(Persson et al. 1996)
Tanzanian (<i>n</i> = 248)	0.82 ^b	0.18 ^b	–	–	(Herrlin et al. 1998)
Zimbabwean (<i>n</i> = 84)	0.87 ^a	0.13 ^a	–	–	(Masimirembwa et al. 1995)
North America					
African American (<i>n</i> = 15)	0.83	0.17	1	0	(Blaisdell et al. 2002)
African American (<i>n</i> = 108)	0.75 ^b	0.25 ^b	–	–	(Goldstein et al. 1997)
Caucasian (<i>n</i> = 24)	0.92	0.08	1	0	(Blaisdell et al. 2002)
European American (<i>n</i> = 105)	0.87 ^a	0.13 ^a	–	–	(Goldstein et al. 1997)
Europe					
Swiss (<i>n</i> = 207)	0.84 ^a	0.16 ^a	–	–	(Crettol et al. 2005)
Asia					
Asian decent (<i>n</i> = 24)	0.69 ^b	0.31 ^b	1	0	(Blaisdell et al. 2002)
Japanese (<i>n</i> = 53)	0.77 ^b	0.23 ^b	–	–	(Goldstein et al. 1997)
Filipino (<i>n</i> = 52)	0.61 ^b	0.39 ^b	–	–	(Goldstein et al. 1997)
Saudi Arabian (<i>n</i> = 97)	0.85 ^a	0.15 ^a	–	–	(Goldstein et al. 1997)

^aSignificant at *p* < .05 when compared with Egyptians.

^bSignificant at *p* < .001 when compared with Egyptians.