Allele and genotype frequencies of polymorphic cytochromes P450 (CYP2C9, CYP2C19, CYP2E1) and dihydropyrimidine dehydrogenase (*DPYD*) in the Egyptian population

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Aims The goal of this study was to determine the frequencies of important allelic variants of *CYP2C9*, *CYP2C19*, *CYP2E1* and *DPYD* in the Egyptian population and compare them with the frequencies in other ethnic populations.

Methods Genotyping of *CYP2C9* (**2* and **3*), *CYP2C19* (**2* and **3*), *c2* variant of *CYP2E1* and *DPYD* alleles (**2 A*-**6*) was carried out in a total of 247 unrelated Egyptian subjects. An allele-specific fluorogenic 5′ nuclease chain reaction assay was applied for detection of *CYP2C9* and *CYP2C19* variants. Other variants of the *CYP2E1* and *DPYD* genes were determined using polymerase chain reaction (PCR)-restriction fragment length polymorphism and allele-specific PCR based assays.

Results *CYP2C9* allele frequencies in 247 Egyptian subjects were 0.820 for *CYP2C9*1*, 0.120 for *CYP2C9*2* and 0.060 for *CYP2C9*3*. For *CYP2C19*, the frequencies of the wild type (*CYP2C19*1*) and the nonfunctional (**2* and **3*) alleles were 0.888, 0.110 and 0.002, respectively. *CYP2C19*3*, which is considered an Asian mutation, was detected in one subject (0.40%) who was heterozygous (**1/*3*). Two subjects (0.80%) were homozygous for **2/*2*, while no compound heterozygotes (**2/*3*) or homozygotes for **3* were detected. For *CYP2E1*, only four subjects (1.70%) had the rare *c2* variant, expressed heterozygously, giving an allele frequency of 0.009. Five variants of *DPYD* were analysed, with no splice sites (**2 A*) or ΔC1897 (**3*) found in this population. The frequencies of other variants were 0.028, 0.115 and 0.090 for **4*, **5* and **6*, respectively.

Conclusions Comparing our data with that obtained in several Caucasian, African-American and Asian populations, we found that Egyptians resemble Caucasians with regard to allelic frequencies of the tested variants of *CYP2C9*, *CYP2C19*, *CYP2E1* and *DPYD*. Our results may help in better understanding the molecular basis underlying ethnic differences in drug response, and contribute to improved individualization of drug therapy in the Egyptian population.

Keywords: CYP, *DPYD*, Egyptians, pharmacogenetics

Introduction

The cytochrome P450 enzymes (CYP) play a central role in the metabolism of many therapeutic agents. Differences in the activities of these enzymes are thought to be responsible for individual variability in drug response and/or toxicity. Among the CYP enzymes many isoforms exhibit genetic polymorphism, examples include 2C9, 2C19 and 2E1.

CYP2C9 catalyses the oxidation of clinically important drugs including phenytoin, tolbutamide, warfarin and a large number of nonsteroidal anti-inflammatory drugs [1]. Previous studies have shown that changes in amino acid composition of this enzyme can affect both its activity and substrate specificity, and thereby produce individual variability in the elimination and/or dosage requirements

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of prototypic CYP2C9 substrates [2, 3]. Eleven CYP2C9 variant alleles have been reported to date [4]; however, only two amino acids substitutions, *CYP2C9**2* (Arg144 to Cys) and *CYP2C9**3* (Ile359 to Leu), are recognized in humans as main CYP2C9 variants [5, 6]. *CYP2C9**2* and *CYP2C9**3* have reduced catalytic activity compared with wild type (*CYP2C9**1*) [7, 8].

CYP2C19 metabolizes several therapeutically important drugs, namely omeprazole, lansoprozole, propranolol, imipramine, mephenytoin, chloroguanide, hexabarbitone and diazepam [9]. Carriers of two CYP2C19 non-functional alleles have a severely impaired capacity to metabolize drugs that are substrates for this enzyme and are hence designated poor metabolizers (PMs). Nine CYP2C19 variant alleles have been reported to date [10]. Among these variants, two alleles with separate mutations have been associated with the defective enzyme in Caucasian and Asian populations. The first is *CYP2C19**2*, which has a mutation in exon 5 causing an aberrant splice site. The other variant allele is *CYP2C19**3* with a point mutation in exon 4 producing a premature stop codon. The most commonly mutated allele is *CYP2C19**2* in both Asians and Caucasian PMs [11]. The second discovered mutation, *CYP2C19**3*, is rare among Caucasian subjects but accounts for the remaining defective alleles in Asian subjects [12].

Human CYP2E1 is involved in the oxidation of drugs such as ethanol, chlorzoxazone, paracetamol and fluorinated anaesthetics [13]. It can be induced by ethanol, and studies have shown an approximately 50-fold individual variability in its expression [14, 15]. In addition, the *c2* mutant allele in the regulatory 5′ flanking region has been associated with higher transcriptional activity [16].

For CYP2C9, CYP2C19 and CYP2E1 the prevalence of non-functional alleles varies substantially in populations of different racial origin. While the frequencies of mutant alleles of these clinically important enzymes have been studied extensively in all major human races, only limited information has been available for the Egyptian population [17].

Aside from the importance of the CYP system in the biotransformation of drugs, the dihydropyrimidine dehydrogenase (DPD) enzyme is of equal importance, especially in cancer chemotherapy. DPD is the initial and rate-limiting enzyme in the metabolic pathway for the catabolism of the pyrimidine bases uracil and thymine [18]. It is also the key enzyme that degrades the structurally related pyrimidine antimetabolite 5-FU. Patients with low DPD activity are characterized by a decreased elimination of 5-FU from plasma associated with severe haematological, gastrointestinal and neurological toxicity, and often death [19]. Among Caucasian populations DPD activity is highly variable and subject to polymorphic regulation of the *DPYD* gene [20]. Few studies on the frequency of DPD deficiency in general populations have been performed, so the population distribution of the variant alleles and their contribution to reduced DPD activity *in vivo* is not well characterized [21]. Moreover, no data has been reported regarding the frequency of the *DPYD* alleles in the Egyptian population.

We report here the allele frequencies of *CYP2C9* (**2* and **3*), *CYP2C19* (**2* and **3*), the *c2* variant of CYP2E1, and *DPYD* allelic variants (**2 A*, **3*-**6*), in a sufficiently large sample of the Egyptian population (247 subjects). To our knowledge it is the largest genotyping study of the Egyptian population reported to date.

Methods

Subjects

The Egyptian population is divided into several cultural groups: Bedouins, Nubians, Berbers, Peasants and Urbanites. The Bedouins and Berbers are small minorities. They are still nomadic tribal people living in isolated oases and roaming through the country's vast region so they rarely communicate or interact with other Egyptian cultural groups. The Nubians are the people who lived for thousands of years in their own land along the Nile, called Nuba, which overlapped from the southern part of Egypt into northern Sudan. The Nubians had their own ancient language and their own unique cultural traits. They are darker in complexion than the rest of the Egyptian people and they have heavier facial features and coarser hair. The Peasants are the people who inhabit the rural villages that line the Nile. For a half century the majority of this rural population has shifted to the main Urban centres in search of employment or education. The Urbanites represent more than half of the Egyptian population and reside the big cities (e.g. Cairo and Alexandria). However, there is a long history of tensions between the Urbanites and the Peasants and many of them inter-married.

Two hundred and forty-seven unrelated Egyptian subjects participated in the present study. All these subjects were students and staff at Cairo University, thereby considered as Urbanites living in Cairo or other surrounding cities. Each subject gave a sample of about 1 ml of saliva after detailed explanation of the purpose of the study; a signed written consent was also obtained from each subject. Genomic DNA was isolated from the saliva using a QIAamp DNA Tissue Kit (QIAGEN, Hilden, Germany) according to the manufacturer's recommendations. Sample collection and DNA isolation were performed under the supervision and ethical approval of the dean of the faculty of pharmacy of Cairo University. The isolated DNA

samples were sent to our laboratory in Japan, and the genotyping protocol was approved by the institutional ethics committee of Tohoku University School of Medicine, Sendai, Japan and Cairo University Faculty of Pharmacy, Cairo, Egypt.

Genotyping

CYP2C9 and CYP2C19

An allele-specific fluorogenic 5′ nuclease chain reaction assay was performed for detecting polymorphisms of CYP2C9 (*CYP2C9*2* and **3*), CYP2C19 (*CYP2C19*2* and **3*), using the ABI PRISM 7700 sequence detection system (Applied Biosystems), according to methods described previously by Hiratsuka *et al.* [22].

CYP2E1

The *RsaI* polymorphism in the 5′ flanking region of the gene was investigated by the method described by Kato *et al.* [23].

DPYD

Three *DPYD* alleles were analysed by a polymerase chain reaction (PCR)-restriction fragment length polymorphism assay (**3*, **4* and **5*) as described previously by Ridge *et al.* [24]. An allele-specific PCR assay was developed to determine the alleles **2* and **6*. In brief, DNA was amplified with 0.67 mM primers **2R (5′-CCATCTTACATGGCACCCATTT-3′) and either **2FW (5′-AGGCTGACTTCCCAGACAACG-3′) or **2FM (5′-AGGCTGACTTCCCAGACAACA-3′) were used in the wild-type specific or mutant specific reactions, respectively. The primers used for detection of **6* were **6R (5′-CTCCATATGTAGTTCGCTTTGCAA-3′) and either **6FW (5′-ATAGGTGGTGCCAATGGCG-3′) or **6FM (5′-ATAGGTGGTGCCAATGGCA-3′). PCR amplification consisted of an initial denaturation step at 94° C for 5 min followed by 30 cycles of denaturation at 94° C for 30 s, annealing at 65° C for 3 s (**2*) and at 62° C for 5 s (**6*), extension at 72° C for 1 min (**2*) and for 30 s (**6*). The final extension step was performed at 72° C for 5 min. The PCR products (245 bp and 116 bp) for **2* and **6*, respectively, were separated on 3% agarose gels.

Statistical analysis

Data were compiled according to the genotype and allele frequencies estimated from the observed numbers of each specific allele. The frequency of each allele in our subjects is given together with the 95% confidence interval. Differences in allele frequencies between Egyptians and other ethnic populations were measured by Fisher exact test. A *P* value below 0.05 was considered statistically significant throughout the population comparisons.

Results

CYP2C9

The frequencies of *CYP2C9* alleles among 247 Egyptian subjects were as follows; *CYP2C9*1*, 0.820 (95% CI, 0.784, 0.852), *CYP2C9*2*, 0.120 (95% CI, 0.091, 0.148) and *CYP2C9*3*, 0.060 (95% CI, 0.041, 0.084). The distribution of *CYP2C9* genotypes is summarized in Table 1.

CYP2C19

The frequencies of *CYP2C19* alleles among the 247 Egyptian subjects were as follows; *CYP2C19*1*, 0.888 (95% CI, 0.861, 0.916), *CYP2C19*2*, 0.110 (95% CI, 0.082, 0.137) and *CYP2C19*3*, 0.002 (95% CI, 0, 0.006). The distribution of *CYP2C19* genotypes is summarized in Table 1.

Table 1 Genotype distribution of the tested variants of *CYP2C9*, *CYP2C19* and *DPYD* in the Egyptian population.

Genotype	n	*Observed frequency percentage (95% CI)*	*Predicted frequency percentage by Hardy–Weinberg law*
CYP2C9 (247 subjects)			
**1/*1*	164	66.43 (60.50, 72.28)	67.24
**1/*2*	47	19.00 (14.13, 23.92)	19.68
**1/*3*	29	11.74 (7.72, 15.75)	9.84
**2/*2*	6	2.43 (0.50, 4.34)	1.44
**2/*3*	0	0 (0)	1.44
**3/*3*	1	0.40 (0, 1.00)	0.36
CYP2C19 (247 subjects)			
**1/*1*	194	78.56 (73.42, 83.66)	78.80
**1/*2*	50	20.24 (15.23, 25.25)	19.60
**1/*3*	1	0.40 (0, 1.20)	0.35
**2/*2*	2	0.80 (0, 2.00)	1.21
**2/*3*	0	0 (0)	0.04
**3/*3*	0	0 (0)	0
DPYD (239 subjects)			
**1/*1*	138	57.73 (51.47, 64.00)	58.81
**1/*2*	0	0 (0)	0
**1/*3*	0	0 (0)	0
**1/*4*	13	5.44 (2.50, 8.30)	4.35
**1/*5*	43	18.00 (13.12, 22.86)	17.64
**1/*6*	30	12.55 (8.35, 16.75)	13.80
**4/*4*	0	0 (0)	0.08
**4/*5*	1	0.42 (0, 1.20)	0.64
**4/*6*	0	0 (0)	0.50
**5/*5*	0	0 (0)	1.30
**5/*6*	13	5.44 (2.50, 8.30)	2.07
**6/*6*	1	0.42 (0, 1.20)	0.81

n, number of subjects. The 95% confidence intervals are given in parentheses.

Table 2 Comparison of allele frequencies of *CYP2C9* reported from different ethnic populations.

Population	n	*Frequency (*P *value* vs *Egyptians)* **1*	**2*	**3*	*Reference*
Egyptians	494	0.820	0.120	0.060	Present study
Caucasians					
American	200	0.860	0.080 (NS)	0.060 (NS)	[6]
British	200	0.790	0.125 (NS)	0.085 (NS)	[5]
Swedish	860	0.819	0.107 (NS)	0.074 (NS)	[8]
Turkish	998	0.794	0.106 (NS)	0.100 (NS)	[25]
Asians					
Japanese	436	0.979	0 (<0.0001)	0.021 (0.0020)	[26]
Korean	1148	0.989	0 (<0.0001)	0.011 (<0.0001)	[27]
Chinese-Taiwanese	196	0.974	0 (<0.0001)	0.026 (NS)	[6]
African-Americans	200	0.985	0.010 (<0.0001)	0.005 (0.0004)	[6]

n, total number of alleles.
Differences in allele frequencies were measured by Fisher exact test.
NS indicates that there were no significant differences ($P > 0.05$).

CYP2E1

For CYP2E1 genotyping, 12 samples were excluded due to unsuccessful PCR amplification. From a total of 235 analysed samples, only four subjects (1.7%) were heterozygous for the rare *c2* allele giving an allele frequency of 0.009 (95% CI, 0, 0.017).

DPYD

Because of failure of the PCR amplification of eight samples, only 239 subjects could be analysed for the five points of mutations in the *DPYD* gene. The distribution of *DPYD* genotypes is summarized in Table 1. Neither **2* nor **3* was detected in the present study. The other three variants were found in frequencies of 0.028 (95% CI, 0.014, 0.044), 0.115 (95% CI, 0.090, 0.148) and 0.090 (95% CI, 0.068, 0.120) for **4*, **5* and **6*, respectively.

These results were in good accordance with the expected genotype distributions of the tested genes, calculated by the Hardy–Weinberg equation (Table 1). The frequencies of the tested mutant alleles of *CYP2C19*, *CYP2C19* and *DPYD* in the Egyptian subjects are given in Tables 2, 3 and 4, respectively, with comparison to other data reported from various ethnic populations.

Discussion

Egypt is unique geographically, as it is located centrally to the three continents of Africa, Europe and Asia. Throughout history, the Greeks, Romans, Arabs, Turks, French and British have all ruled Egypt and mixed with its people, such that modern Egypt now is an amalgam of all these legacies. Because of the clear heterogeneity and genetic admixture in the Egyptian population, we considered it especially important to investigate the allele frequencies and genotype distributions of some variants of a pharmacogenetic interest in this population. This study was carried out in 247 unrelated Egyptian subjects, and is among the first to describe many allelic variants in this population.

CYP2C9

The frequency of the *CYP2C9*2* allele in the Egyptian population was 0.120, which is in a range comparable with other Caucasian populations; 0.080 in American [6], 0.125 in British [5], 0.107 in Swedish [8] and 0.106 in Turkish [25]. In the study of Sullivan-Klose *et al.* [6], it was reported that the *CYP2C9*2* allele occurs at a significantly lower frequency in the African-American population (0.010). On the other hand, the *CYP2C9*2* allele was reported to be absent or at least very rare in the East Asian populations [6, 26, 27] (*P* values are listed in Table 2).

The *CYP2C9*3* allele occurred with a frequency of 0.060 in the Egyptian subjects. This finding is similar to that of other Caucasian populations in which the frequency of the *CYP2C9*3* allele was reported to be 0.060 in American [6], 0.085 in British [5], 0.074 in Swedish [8] and 0.100 in Turkish [25]. Lower frequencies were reported for the *CYP2C9*3* variant in East Asian populations (0.011–0.021) in Japanese [26], Korean [27] and Chinese-Taiwanese [6]. The lowest frequency of *CYP2C9*3* was reported in African-Americans (0.005) [6] (*P* values are listed in Table 2).

The catalytic activity of the *CYP2C9*3* encoded enzyme is much lower than those of *CYP2C9*1* and **2* [28]. Moreover, the clearance of orally ingested S-warfarin *in vivo* was reduced by 66% and 90% among

Table 3 Comparison of allele frequencies of *CYP2C19* reported from different ethnic populations.

Population	n	*Frequency (*P *value* vs *Egyptians)* *1	*2	*3	*Reference*
Egyptians	494	0.888	0.110	0.002	Present study
Caucasians					
American	210	0.871	0.129 (NS)	0 (NS)	[35]
Canadian (Inuit)	304	0.890	0.110 (NS)	0 (NS)	[36]
Danish	478	0.839	0.161 (0.0200)	0 (NS)	[37]
Swedish	166	0.849	0.144 (NS)	0.007 (NS)	[38]
German	280	0.850	0.150 (NS)	0 (NS)	[39]
Portuguese	306	0.870	0.130 (NS)	0 (NS)	[40]
Australian	198	0.854	0.146 (NS)	0 (NS)	[41]
Saudi Arabia	194	0.850	0.150 (NS)	0 (NS)	[35]
Asians					
Japanese	106	0.670	0.230 (0.0022)	0.104 (<0.0001)	[35]
Korean	206	0.675	0.209 (0.0008)	0.116 (<0.0001)	[42]
Chinese-Taiwanese	236	0.630	0.320 (<0.0001)	0.055 (<0.0001)	[35]
Filipino	104	0.540	0.390 (<0.0001)	0.077 (<0.0001)	[35]
South-west Asia					
North Indian	242	0.700	0.300 (<0.0001)	0 (NS)	[43]
Africans					
Bantu-Tanzanian	502	0.815	0.179 (0.0021)	0.006 (NS)	[42]
Ethiopian	228	0.846	0.136 (NS)	0.018 (0.0400)	[44]
Venda	304	0.783	0.217 (<0.0001)	0 (NS)	[45]
Zimbabwean	336	0.869	0.131 (NS)	0 (NS)	[45]
African-Americans	216	0.750	0.250 (<0.0001)	0 (NS)	[35]

n, total number of alleles.
Differences in allele frequencies were measured by Fisher exact test.
NS indicates that there were no significant differences ($P>0.05$).

Table 4 Comparison of allele frequencies of *DPYD* reported from different ethnic populations.

Population	n	*Frequency of DPYD variants (*P *value* vs *Egyptians)* *2 A†	*3‡	*4	*5	*6	*Reference*
Egyptians	478	0	0	0.028	0.115	0.090	Present study
Caucasians							
Finish	180	0.011	n.d.	0.033 (NS)	0.072 (NS)	0.067 (NS)	[47]
British	120	0	0	0.008 (NS)	0.280 (<0.0001)	0.058 (NS)	[24]
Asians							
Japanese	100	0	n.d.	0.011 (NS)	0.350 (<0.0001)	0.044 (NS)	[47]
Taiwanese	262	0	n.d.	0 (0.0058)	0.210 (0.0008)	0.014 (<0.0001)	[47]
African-Americans	210	0	n.d.	0.005 (NS)	0.227 (0.0002)	0.019 (0.0004)	[47]

n, total number of alleles. n.d., not determined.
Differences in allele frequencies were measured by Fisher exact test.
NS indicates that there were no significant differences ($P>0.05$).
†, no significant differences had been observed between the Egyptians and the other populations.
‡, no significant difference had been observed between the Egyptians and the British populations.

subjects heterozygous and homozygous for the *CYP2C9*3* allele, respectively, compared with homozygous *CYP2C9*1* subjects [29]. In our study only one subject was homozygous for **3/*3* (0.40%), which is in a range comparable with British (0.10%) [5], Swedish (0.70%) [8] and Turkish (0.80%) [25]. On the other hand, no subjects homozygous for *CYP2C9*3* were identified in Eastern populations [6, 26, 27].

CYP2C19

The genetic polymorphism of CYP2C19 has been shown to have the most striking interethnic variation of a CYP so far. The PM frequency ranges from 2 to 7% in Caucasians, 14–25% in Asians, and 60% in the Vanuatu [30, 31]. It has been shown that a higher concentration of omeprazole in PMs results in great gastric acid suppression as compared with extensive metabolizers (EMs) [32]. Cure rates for *Helicobacter pylori* in patients receiving omeprazole and amoxicillin were found to be 28% in homozygous EMs (*CYP2C19*1/*1*), 60% in heterozygous EMs (*CYP2C19*1/*2* and **1/*3*), and 100% in PMs (*CYP2C19*2/*2* and **2/*3*), indicating the importance of dose adjustment in the case of EMs [33].

CYP2C19 genotyping was conducted in 1356 Caucasian individuals throughout the world [34]. In the present study, we found that the incidence of *CYP2C19*2* in Egyptians (0.110) was similar to that found in other Caucasian populations (0.110–0.161) residing in America [35], Canada [36], Europe [37–40], Australia [41] and Saudi Arabia [35]. Higher frequencies of *CYP2C19*2* were reported for Asian populations (0.209–0.390) [35, 42], North Indians (0.300) [43], Africans (0.131–0.217) [42, 44, 45] and African Americans (0.250) [35] (*P* values are listed in Table 3).

On the other hand, the discovered presence of *CYP2C19*3* in one Egyptian subject (0.002) was an interesting finding because limited numbers of studies have reported *CYP2C19*3* in populations other than the Asians. *CYP2C19*3* was reported in the Swedish population (0.003) [32], (0.007) [38], the Bantu-Tanzanian (0.006) [42] and the Ethiopian (0.018) [44] (*P* values are listed in Table 3). *CYP2C19*3* has been regarded as an Asian mutation and accounted for the remaining alleles in Asian PMs after genotyping for *CYP2C19*2*. An explanation for our finding of *CYP2C19*3* in the Egyptian subjects may be the emigration and possible genetic admixture between the Egyptians and Asian populations.

CYP2E1

Significant interethnic difference exists in the *CYP2E1* polymorphism among different populations [23, 46]. The frequency of the mutant *c2* allele is about 20% in Japanese, while it is present only in 2–4% of Caucasians and 1–2% of African-Americans [23]. The frequency of the *c2* allele was previously detected in 42 Egyptian subjects (0.010) by Anwar *et al.* [17]. In the present study we obseved a frequency of 0.009 of the *c2* allele by analysing a larger number of Egyptian subjects confirming the rare occurrence of this mutant allele in the Egyptian population. This rare occurrence may prove that any influence of the *CYP2E1* gene on diseases such as cancer or alcoholic liver disease, or on hepatotoxicity following the use of halogenated anaesthetics is likely to be minimal in this population.

DPYD

Polymorphic drug metabolizing enzymes often have a limited number of variant genotypes that account for most individuals with reduced or deficient phenotypes. In the case of DPD, although 13 variant *DPYD* alleles have been identified to date in DPD-deficient subjects, the majority of these alleles are not associated with the DPD deficiency [21]. However, because of the severity of 5-FU induced toxicity, it would be advantageous to prescreen cancer patients for levels of DPD activity and/or *DPYD* genotype before 5-FU therapy and modify the dose appropriately. The present study was the first aimed to detect the frequencies of five *DPYD* variants in the Egyptian population and compare them with those of other ethnic populations.

No exon 14 splice site mutations (*DPYD*2 A*) were found among the 239 subjects studied. This allele was not found among 60 British [24], 50 Japanese, 131 Taiwanese, or 105 African-Americans [47], but had a 0.011 frequency in the Finnish population [47]. Previous studies [47–49] demonstrated that intron 14 G1A (*DPYD*2 A*) results in the production of a truncated mRNA, and has been consistently associated with low DPD activity and 5-FU toxicity. However, one patient with a heterozygous intron 14 G1A mutation had normal DPD activity [21]. This fact was explained by the possible allelic regulation of the *DPYD* gene that may occur to allow a normal DPD phenotype through increased expression of the wild-type allele [21].

No subject with ΔC1897 (*DPYD*3*) was detected. This allele results in premature termination of translation prior to the uracil binding site, and no DPD activity was detectable in a homozygous mutant individual [50]. However, other studies suggested that ΔC1897 is a relatively rare allele in Caucasian populations [21, 24].

The presence of a G to A transition at codon 534 (*DPYD*4*) was detected with a frequency of 0.028. This is similar to that found in Finnish (0.033) [47], British (0.008) [24], Japanese (0.011) and African-Americans (0.005) [47]. In the study of Ridge *et al.* [24], one individual with a codon 534 mutation had the lowest DPD activity, whereas two previously described colorectal cancer patients with codon 534 mutations had DPD activities greater than the average value [51], making the impact of this mutation unclear.

Mutations at codon 543 (*DPYD*5*) were found with a frequency of 0.115, also similar to that found in the Finnish subjects (0.072) [23]. However, this allele was

more common in British (0.280) [22], Japanese (0.350), Taiwanese (0.210) and African-Americans (0.227) [23] (*P* values are listed in Table 4). This mutation was associated with a wide range of mononuclear cell DPD activity [22, 51], suggesting that this mutation is a common polymorphism, and alone is not associated with impaired DPD activity.

Mutations at codon 732 (*DPYD*6*) were also a relatively common finding (0.090). This is similar to that found in Finnish (0.067) [47], British (0.058) [24] and Japanese (0.014) [47] but is more frequent than that found in Taiwanese (0.014) and African-Americans (0.019) [47] (*P* values are listed in Table 4). This mutation was also found in individuals with a wide range of DPD activity [21, 24], confirming that it is not a functional mutation.

In the present study, 15 subjects (6.7%) had more than one mutation in the *DPYD* gene. In the study of Ridge *et al*. [24], the three individuals having more than one mutation in the *DPYD* gene had DPD activity values lower than 10% of the population, suggesting that accumulation of *DPYD* mutations may contribute to low activity phenotype of the enzyme.

In conclusion, the tested allelic variants of *CYP2C9*, *CYP2C19*, *CYP2E1* and *DPYD* existed in Egyptian people with frequencies comparable to other Caucasian populations with some differences among Asians and African Americans. It is hoped that our results will aid in understanding the ethnic diversity of the Egyptian population, and offer a preliminary basis for more rational use of drugs that are substrates for CYP2C9, CYP2C19, CYP2E1 and DPD in this population.

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