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Difference in Frequencies of the Cystic Fibrosis Alleles, ΔF508 and W1282X, between Carriers and Patients

Key Words
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Abstract

One major mutation, ΔF508, causing cystic fibrosis (CF) is found in most populations around the world. Among CF patients of Jewish Ashkenazi origin two major mutations, W1282X and ΔF508 were found. We compared the relative frequencies of the two major mutations found in this patient population to their relative frequencies in the healthy population. The studied patient population included the entire CF Jewish Ashkenazi patient population in Israel (238 chromosomes), and a small group of Jewish Ashkenazi patients in the USA (57 chromosomes). Among these, 79 (27%) chromosomes carried the ΔF508 mutation, and 151 (51%) the W1282X mutation. In addition, we have analyzed the results of screening 1,946 unrelated healthy Jewish Ashkenazi individuals for the ΔF508 and the W1282X mutations. Surprisingly, an almost equal number of carriers of the ΔF508 (35) and W1282X (36) was found. The difference between the relative proportions of the mutations in the two groups is statistically significant ($p = 0.025$). A striking manifestation of this difference is revealed in the analysis of patients' genotypes. There were 36 patients homozygous for W1282X, while only 7 patients were homozygous for ΔF508, although the number of ΔF508 carriers in the general Jewish Ashkenazi population is almost equal to the number of W1282X carriers. This difference in allele frequencies found between healthy carriers and CF patients in the Jewish Ashkenazi population might not be unique to this ethnic group nor to the CF disease. The results indicate that the common practice of inferring general population epidemiologic parameters directly from patients information is liable to introduce biases. Thus, widespread screenings should be preceded by smaller-scale pilot studies designed to verify that the mutation frequencies in the patient population represent the carrier frequencies in the relevant general population.

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Introduction

Cystic fibrosis (CF) is the most common lethal recessive inherited disease in the Caucasian population [1]. The identification of the CF gene revealed a major mutation causing the disease, ΔF508, which was found in about 70% of the CF chromosomes around the world [2]. About 300 additional, much less frequent, mutations were later identified by the International CF Genetic Analysis Consortium. The frequencies of CF mutations vary substantially between patients from different ethnic groups. A previous study on a relatively small number of the Jewish Ashkenazi CF patient population in Israel revealed a major mutation, W1282X, which accounts for 60% of the CF chromosomes in this ethnic group [3]. ΔF508 accounts for an additional 23% of the CF chromosomes. Altogether, six mutations identify 95% of the mutant alleles in CF chromosomes of Jewish Ashkenazi patients [3, 4]. The very high proportion of characterized CF alleles in this patient population sparked carrier-screening studies in the general healthy Jewish Ashkenazi population in Israel and the United States. The aim of this study was to compare the relative frequency of the W1282X and the ΔF508 mutations among the CF Jewish Ashkenazi patient population to their relative frequency among healthy carriers of Jewish Ashkenazi origin.

Patients and Methods

The patients analyzed in the present study attended the CF clinics at Chaim Shiba Medical Center, Tel Aviv, Shaare Zedek Medical Center, Jerusalem, Soroka Medical Center, Beersheva, Carmel Medical Center, Haifa, Rambam Medical Center, Haifa, Belinson Medical Center, Petach Tikva, Hadassah Medical Center, Jerusalem, and the Baylor College of Medicine, Houston, Tex., USA. The patients' diagnoses had been previously confirmed by typical clinical findings of pulmonary and/or gastrointestinal disease [1]. The ΔF508 and the W1282X mutations were analyzed as described previously [3, 5]. CF screenings in Israel were carried out in two centers. The Chaim Shiba Medical Center, and the Haddassah Medical Center. The CF screen in the United States was carried out at the Mount Sinai Medical Center, New York. In all studies, only healthy unrelated individuals (most of whom were adults) of Jewish Ashkenazi origin, with no known history of CF in their families were analyzed.

The significance of the difference between the relative allele proportions was calculated using the test of difference between two proportions [6].

Results

A total of 238 obligate Jewish Ashkenazi CF chromosomes, from CF patients, from all the CF centers in Israel were included in the study. These chromosomes represent 95% of the chromosomes carried by the entire Jewish Ashkenazi patient population in Israel, including chromosomes from known patients who are no longer alive. Among these, 116 (49%) chromosomes were found to carry the W1282X mutation, and 64 (27%) chromosomes were found to carry the ΔF508 mutation. These results, from the entire Israeli population, verify that W1282X is the major mutation among Jewish Ashkenazi CF patients. The analysis was extended by analyzing 57 American CF chromosomes of Jewish Ashkenazi origin. Similar frequencies were found in this patient population. There were 15 (26%) chromosomes with the ΔF508 mutation, and 35 (61%) with the W1282X mutation (table 1). Therefore, we combined mutation frequencies found among the Israeli and American Jewish Ashkenazi CF chromosomes (table 1). It can be seen that the most common mutation in the general world population, ΔF508, is far less abundant in the general Ashkenazi CF patient population (27%). The frequency of the major mutation in this patient population, W1282X, is almost twice (51%) the frequeny of the ΔF508 mutation.

Table 1. CF alleles in chromosomes carring the ΔF508 and W1282X mutations

	Number of chromosomes	W1282X	ΔF508	$\frac{\Delta F508}{\Delta F508 + W1282X}$	Source
CF alleles in patients					
Israel	238	116	64	0.36	
USA	57	35	15	0.30	
Total	295	151	79	0.34	
CF alleles in the healthy population					
Israel	1,874	17	15	0.47	Abeliovich [pers.commun.]
	618	7	7	0.50	Zamostiano et al. [7]
USA	1,400	12	13	0.52	Eng [pers. commun.]
Total	3,892	36	35	0.49	

We have collected and analyzed data from three independent screening studies of unrelated individuals carried out in Israel and the USA (table 1). Among the 1,946 Ashkenazi Jews screened, an essentially equal number of ΔF508 and W1282X carriers was independently found in each study. In total, 35 carriers of ΔF508 and 36 carriers of W1282X were found. The relative proportion of ΔF508 [ΔF508/(ΔF508+W1282X)] estimated from the general population was 0.49, whereas the relative proportion estimated from the CF patients was 0.34. The difference in the proportions, calculated using the test of difference between two proportions [6], was statistically significant ($p = 0.025$). A striking manifestation of this difference is presented in table 2. It can be seen that although the number of ΔF508 carriers in the general Jewish Ashkenazi population is almost equal to the number of W1282X carriers, there were 36 patients homozygous for W1282X, while only 7 patients were homozygous for ΔF508. In order to test whether there is also a difference in heterozygous patients carrying ΔF508, we analyzed the frequency of the ΔF508 mutation in compound-heterozygote patients whose other CF allele was not W1282X, and the frequency of the W1282X mutation in compound heterozygote patients whose other CF allele was not ΔF508. As can be seen in table 2, 42 such patients were found. The relative proportion of the ΔF508 allele in this group of patients is 0.43. This relative proportion was not significantly different from either the relative proportion in carriers or patients.

Table 2. Genotypes of CF patients

Genotypes	Israel	USA	Total
W1282X/W1282X	25	11	36
ΔF508/ΔF508	6	1	7
W1282X/ΔF508	31	7	38
W1282X/other[1]	22	2	24
ΔF508/other[2]	15	3	18
Other/other	5	0	5

Patients were included in this analysis only if both parents were of Jewish Ashkenazi origin.

[1] Compound heterozygous patients carrying the W1282X allele whose other allele is not ΔF508.

[2] Compound heterozygous patients carrying the ΔF508 allele whose other allele is not W1282X.

Discussion

This study revealed a significant difference in the relative proportion of two mutant alleles between CF patients and healthy carriers. It is important to note that in the two studied patient populations and in each of the three screening populations, similar relative proportions were found. Thus, the discrepancy seen between patients and carriers was not an unusual phenomenon in one subpopulation. It is also important to note that the Ashkenazi Jews residing in Israel and the USA have a common origin. The results of the study indicate that the common practice of inferring the frequencies of mutant alleles in a general population using the frequencies found in the patient population might not be justified.

This difference, if confirmed by further screens, might not be unique to this ethnic group nor to the CF disease. The fact that the Jewish Ashkenazi CF population carries *two* major mutations enabled us to see this discrepancy. In non-Jewish populations, several CF screening programs have already been initiated, but since in most of these populations there is only one major mutation and many other rare mutations, only a few chromosomes carrying mutations other than ΔF508 were found [8, 9]. Relative-proportion comparisons in these studies could not yield statistically significant results. Further screening studies of a large number of individuals for a large number of mutations are required before any generalization of this phenomenon can be asserted. It is important to note that another difference between CF patients and healthy carriers has already been reported [10]. In that study, a higher than expected frequency of the R117H mutation was found among healthy carriers. Here also, relative allele frequencies found in CF patients were different from the relative proportions in the screened healthy population. This was probably due to a milder expression of the disease in patients carrying this mutation.

The estimated mutant allele frequency in a population is used to calculate epidemiologic parameters such as carrier frequencies [11]. These parameters are mainly used in genetic counselling, in particular to calculate the residual risk of counselled couples, where at least one of them has not been identified as a carrier. In addition, these estimates are used to calculate the cost-effectiveness of proposed screening programs. Any significant bias, such as that found in this study in the Jewish Ashkenazi population, could result in considerable inaccuracies, which might mislead counselled couples and health care policy makers. Since the common practice of inferring general population epidemiologic parameters directly from patient information is liable to introduce biases, we suggest that widespread screenings should be preceded by smaller-scale pilot studies designed to verify that the mutation frequencies in the patient population represent the carrier frequencies in the relevant general population. Until this is accomplished, calculations of general population frequencies and risk based on frequencies of identified mutations in identified patients, should be treated with caution.

Various mechanisms can lead to this marked difference between the relative mutant allele frequencies. Since our data do not indicate any nonrandom mating it is plausible that the difference is a result of postzygotic (postnatal or prenatal) or prezygotic advantage or disadvantage of a certain allele or genotype. First, the mechanism involved might be postnatal selection against homozygotes for the ΔF508 mutation. Both the ΔF508 and the W1282X mutation were found to be associated with the same severe form of the disease [3, 12, 13]. But since the analyzed patient population comprised patients of dif-

ferent ages, it is impossible to exclude the possibility that the bias is a result of unidentified patients homozygous for the ΔF508 mutation, who died postnatally. Thus, studies aimed at verifying the survival of patients of Ashkenazi origin carrying the ΔF508 and/or the W1282X mutations are required for testing this hypothesis. The results of such studies will contribute to our understanding the phenotype-genotype correlation of the ΔF508 mutation. It is important to note that the proportion of ΔF508 carriers in the general Jewish Ashkenazi population, found in the abovementioned screening programs (1.8%, see table 1), is close to that found in general Caucasian populations (2.2–3.1%) [8, 9, 11]. If there is no a priori reason to suppose that ΔF508 should be less abundant in the Jewish Ashkenazi population, then selection against homozygotes for ΔF508 in this population might explain the findings. This possibility was tested by analyzing the ratio between the frequency of the ΔF508 mutation in compound heterozygote patients whose other CF allele is not W1282X, and the frequency of the W1282X mutation in compound heterozygote patients whose other CF allele is not ΔF508. As can be seen in table 2, the number of such patients was too small and thus this analysis resulted in a nonconclusive answer.

Another possible postzygotic (pre- or postnatal) mechanism that might explain the observed difference, is selection against homozygotes for a disadvantageous recessive allele, in linkage with ΔF508 in the Jewish Ashkenazi population. Such a mechanism might also explain the low frequency of the otherwise common mutation ΔF508 in certain populations, including Ashkenazi Jews.

A prezygtoic effect could be a result of non-Mendelian transmission of alleles from carrier parents to their offspring. Such a transmission has already been implicated in a CF study by Williams et al. [14], in which single sperm from a healthy carrier of the ΔF508 mutation were individually genotyped. The study revealed a small, yet statstically significant, difference between the proportion of sperm carrying the ΔF508 mutation (44%), and the proportion of sperm carrying the normal allele (56%). Such a finding needs further validation, but it raises the possibility that meiotic drive, an effect already described in the mouse and fruit fly [15], might occur in humans too. Another mechanism that can cause the difference between allele frequencies found in healthy carriers and patients is transmission bias, as recently discovered in Siberian mice [16]. According to this model, oocyte fertilized by a sperm carrying the W1282X allele, would tend to exclude the normal allele from the embryo. It is also possible that such a mechanism would act against homozygosity for the ΔF508 allele.

In summary, the results of this study indicate a difference in the frequencies of two CF mutations between patients and healthy carriers. The results indicate that the common practice of inferring general population epidemiologic parameters directly from patient information might not be justified.

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Note Added in Proof

Recently, in two human diseases myotonic dystrophy and cone-rod retinal dystrophy, both mapped to 19q13, unexpected segregation distortion has been found [Evans et al., Carey et al.: Nature Genet 6: February 1994]. In the studied families preferential transmission of the mutant alleles from parents to their offspring was found. The mechanism suggested in both studies is meiotic drive.