Mutation Distribution in Expanded Screening for Cystic Fibrosis: Making Up the Balance in a Context of Ethnic Diversity

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Cystic fibrosis (CF) (OMIM 219700) is a serious autosomal recessive condition that primarily affects the respiratory, gastrointestinal, and reproductive organ systems. Its pathophysiology is due to dysfunctional regulation of chloride ion channels precipitated by mutations in the CFTR [cystic fibrosis transmembrane conductance regulator (ATP-binding cassette subfamily C, member 7)] gene (OMIM *602421). CF is typically associated with considerable morbidity and a reduced life expectancy, although phenotypic variability is the norm. This variation is largely due to differences in the combination of underlying molecular defects, but it also stems from the effects of modifying genes, the environment, and complications (1). At one end of the phenotypic spectrum, affected individuals may have very mild manifestations that are isolated to single organ systems, whereas the disease at the other end involves an early demise (2). CF affects populations across the globe but is most common in northern European whites (approximately 1 in 2500 individuals) and in Ashkenazi Jews (approximately 1 in 2270) (3–5). In the ethnically diverse population of the US, CF is diagnosed in approximately 1 in 3900 individuals (6).

The Cystic Fibrosis Genetic Analysis Consortium database includes 1865 CFTR sequence variants (7). Of these variants, p.F508del is the most common mutation; it accounts for nearly two-thirds of the mutations overall. The remaining alleles, many of which are rare, are widely distributed, predominantly in the coding sequence, and demonstrate striking heterogeneity. Both the spectrum and the frequency of individual CFTR sequence changes vary considerably among different ethnic/geographic populations, although much remains to be learned about CFTR mutations in US minorities (8, 9). This situation primarily reflects the fact that these populations have not yet been studied as thoroughly as whites; however, it may also be due in part to the possibility that mutations not typically identified by sequencing methods are more prevalent in one or more minority populations and therefore have not yet been appreciated. For example, although the frequency of large rearrangements is thought to approximate 3% of all affected CFTR alleles, rearrangements are considerably more frequent in some populations (10). Whether a panel test or more-comprehensive testing methods are used, the inclusion in CF tests of mutations that are disproportionately present in non-white, non-Ashkenazi Jewish ethnicities would be expected to produce better diagnostic sensitivities in these and mixed populations. Splice site variant c.2988+1G>A (legacy name, c.3120+1 G>A, intervening sequence 16) illustrates this concept well. Although it is not among the most prevalent mutations in the US, this mutation is the second most prevalent CF allele known in blacks, with a relative frequency of at least 12.2% (11). Thus, a single but comparatively rare mutation overall can have a large effect on mutation detection in the black population. The increased frequency of certain alleles in different ethnic populations is an important consideration in the design of a screening program or diagnostic test.

In 2001, the American College of Medical Genetics (ACMG) and the American College of Obstetricians and Gynecologists (ACOG) proposed a new standard of care with the implementation of prenatal CF carrier screening. This new standard of care recommended offering testing to the non-Hispanic Caucasian and Ashkenazi Jewish populations and to make it available to other racial/ethnic groups (4, 12). After many years of careful consideration, only mutations with an allele frequency of ≥0.1% among the general affected US population were recommended for population-wide CF screening. The decision in 2004 to reduce the original panel of 25 mutations to 23 (13) was based on a lower actual frequency of 1 mutation (c.948delT; legacy name, c.1078delT in exon 7) and the lack of pathogenicity of another (p.N147T). The panel was kept relatively small because of the economic and practical considerations associated with a universal population-screening program and because expanded panels would provide minimal additional yield in the general

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2 Nonstandard abbreviations: CF, cystic fibrosis; ACMG, American College of Medical Genetics; ACOG, American College of Obstetricians and Gynecologists.

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population (4). With the implemented panel, however, differences between individual ethnic groups in detection rates do exist. For example, the detection rate among Hispanic CFTR mutation carriers is approximately 57%, compared with approximately 80%–97% in other whites (4), whereas the Hispanic carrier frequency of 1 in 46 is only modestly lower than that in Caucasians (1 in 29) (4). Although the currently recommended panel is arguably imperfect, the addition of mutations does not necessarily culminate in a superior panel with an appreciably improved detection rate. Expanded panels may create a false sense of security, especially if mutations were arbitrarily chosen with respect to frequency and genotype–phenotype correlations. Conversely, such panels may lead to a false sense of danger when individuals to be screened think that a larger panel would significantly increase test sensitivity even when they are, for example, of northern European origin (14). Since the original implementation of CF carrier screening, the test has been widely implemented across the US, and a plethora of laboratory-developed and commercial assays that contain a minimum of the 23 mutations did demonstrate a moderate increase in mutation detection in minority groups and added value to the screening in these populations. This finding is further substantiated by the observation that the difference between ethnic groups in mutation distribution was statistically significant. Nineteen of the identified mutations were seen in just a single ethnic group. Among Caucasians, the 23-mutation panel covered 90.7% of mutations identified. The remaining 9.3% were distributed over 44 additional mutations on the panel. In comparison, use of the 23-mutation panel for blacks, Hispanics, and Asians covered only 77.3%, 73.1%, and 63%, respectively, of all identified mutations. Overall, 1 in every 38 individuals was a carrier.

Another large study of carrier-screening data was recently published in Genetics in Medicine (16). Direct comparisons of the 2 publications are limited by the different study purposes and cohorts, but some observations can be made. The main purpose of the study by Strom et al. (16) was to determine performance, based on the observed vs predicted detection rates, of the 23-mutation panel recommended by ACMG/ACOG. The authors concluded that the panel is performing as expected. They identified a carrier frequency of 1 in 38.4. For the individuals for whom ethnic background could be obtained (1 192 353 of 2 975 649 individuals), approximately 35% were black, Hispanic, or Asian, and 65% were white or Ashkenazi Jewish. The indication for testing, however, was not captured. The cohort largely comprised individuals referred for carrier screening, but it also included affected individuals and those with a family history of CF. Strom et al. raise the point that expanded panels frequently include mutations of unvetted clinical significance, such as p.D1152H. Assignment of clinical impact indeed continues to be a challenge for certain mutations in any CF test, especially missense mutations of relatively low frequency for which functional studies have not been performed and clinical correlations are not obvious. There may be value in testing for such sequence changes, however, in part because they could then be studied in the context of symptoms, but that depends on the purposes for which the panel is used. For carrier screening per se, a focus on pathogenic mutations associated with substantial morbidity is preferable, given the potential consideration of pregnancy termination when 2 sequence changes are identified. Deserving mention, however, is that the original panel of 25 mutations was based on a frequency of \( \geq 0.1\% \), regardless of whether they had been associated with mild or severe disease (4). Interestingly, the use of expanded genotyping does offer the possibility of identification of sequence changes that occur at a frequency higher than originally appreciated. For example, the new frequency data of Rohlf et al. (15) suggest that c.54-5940_273+
10250del21kb (legacy name, CFTR dele2,3) has a carrier frequency much higher than originally reported.

In conclusion, the 23-mutation panel that reflects the current standard of care appears to adequately meet the original expectations after having been widely implemented and used for several years. Even so, the debate about the value of expanded panels is certain to continue for some time to come. To assess the utility of such panels requires that the application(s), composition, and populations to be studied be taken into account. Knowledge of mutation variety and frequency is still limited for non-Hispanic nonwhite populations, and this fact restricts the sensitivities for mutation detection in any panel-based test. Large studies, such as the one described in this issue of Clinical Chemistry, however, provide valuable insights into the significance of a subset of mutations that are present in the ethnically diverse population of the US.

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References