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Improved detection of cystic fibrosis by the California Newborn Screening Program for all races and ethnicities

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Abstract

Background: Newborn screening (NBS) for cystic fibrosis (CF) is universal in the United States. Protocols vary but include an immunoreactive trypsinogen (IRT) level and *CFTR* variant panel. California CF NBS has a 3-step screening: IRT level, variant panel, and *CFTR* sequencing if only one variant identified on panel.

Methods: This was a cohort study of infants with CF born in California (2007–2021) to examine racial and ethnic differences in having a false-negative NBS result for CF and at which step the false-negative occurred. We examined how different *CFTR* variant panels would improve detection of variants by race and ethnicity: original 39-variant panel, current 75-variant panel, and all 402 disease-causing *CFTR* variants in the CFTR2 database.

Results: Of the 912 infants born in California with CF, 84 had a false-negative result: 38 due to low IRT level and 46 with a high IRT value (but incomplete variant detection). Asian (OR 6.3) and Black infants (OR 2.5) were more likely to have a false-negative screening result than non-Hispanic white infants. The majority of false-negative screening (but CF diagnosis) cases among American Indian/Native Alaskan and non-Hispanic White infants were due to low IRT

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AUTHOR CONTRIBUTIONS

Meghan E. McGarry: Conceptualization; writing—original draft; writing—review and editing; investigation. **Stanley Sciortino:** Formal analysis; writing—review and editing; investigation. **Steve Graham:** Formal analysis; writing—review and editing; investigation. **Tracey Bishop:** Supervision; writing—review and editing; investigation. **Elizabeth R. Gibb:** Conceptualization; investigation; writing—original draft; writing—review and editing.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

levels. The majority of Asian and Hispanic infants with false-negative screening had no variants detected. Detection of two *CFTR* variants was improved with the 75-variant panel in Black, Hispanic, and non-Hispanic White infants and with the 402-variant panel in Black, Hispanic, non-Hispanic White, and other race infants.

Conclusions: Larger *CFTR* panels in NBS improved the detection of CF in all races and ethnicities.

Keywords

cystic fibrosis; genetic diversity; genetic variants; health equity; newborn screen

1 | INTRODUCTION

Since 2010, there has been universal screening for cystic fibrosis (CF) in newborn screening (NBS) programs in the United States. NBS for CF was introduced in Colorado in 1982 as a pilot project using an immunoreactive trypsinogen (IRT) assay.[1] IRT, a precursor of trypsin, can be elevated in infants with CF when pancreatic enzyme release is impaired and the precursor is not converted to an active form and adequately removed from the blood.[2,3] All CF NBS in the United States start by measuring an IRT level and, if elevated, proceed with a second tier of testing. Initially, some CF NBS protocols were IRT/IRT,[4] but due to the high rate of false-negative results, all CF NBS protocols in the United States now include a *CFTR* variant panel as the second-tier test. CF NBS protocols vary greatly among states, including the number of IRT measurements and the number of *CFTR* variants on the screening panel from one to over 300 variants.[5] New York, California, and Wisconsin CF NBS protocols all include sequencing of *CFTR* rather than just a screening panel of *CFTR* variants.[6–8] California introduced in 2007 a 3-step NBS protocol for CF: first, an IRT measurement in all infants; second, in the top 1.6% of IRT values, a *CFTR* variant panel; third, if only one variant identified on the panel, Sanger sequencing of *CFTR*.[6]

Early diagnosis of CF via NBS has been associated with improved outcomes, including improved nutritional status, a decreased need for treatment, and decreased mortality.[9–13] Despite NBS, patients are still being missed on NBS with false-negative results, leading to delays in diagnosis.[14] Children with false-negative results had a later age at diagnosis, a higher rate of hospital admissions per year, worse lung function, higher rates of *Pseudomonas aeruginosa* isolation, and shorter stature.[15]

Compared to non-Hispanic white infants, Asian, Black, Hispanic or other race infants with CF are more likely to be missed on NBS, known as a false-negative NBS, due to the *CFTR* variants included on NBS variant panels.[16] People of different races and ethnicities have varying frequency of *CFTR* variants, and some *CFTR* variants only occur in certain races and ethnicities[17,18] In 2017, California analyzed and compared the effects of expanding the American College of Medical Genetics (ACMG) European ancestry-focused panel to the California 39-variant panel and to a full list of disease-causing variants from CFTR2.org in 2022[19] and demonstrated that additional variants helped better identify newborns of color with CF. The Genetic Disease Screening Program (GDSP) later expanded to a 75-variant panel in 2020.

Given California is one of the most racially and ethnically diverse states in the United States,[20] we examined how the California NBS for CF performed in all races and ethnicities. Incorporating data from all infants born in California since NBS was initiated in 2007, we investigated the rate of false-negative CF NBS results by race and ethnicity and examined the steps leading to the missed screen.

2 | METHODS

2.1 | California CF NBS algorithm

The algorithm of the CF NBS in California is a 3-step process (Figure 1).[6] There is a one-time blood collection via a heel stick after 12 h of age to create dried blood spots (DBSs). The first step of the CF NBS algorithm is a one-time IRT level. DBSs with IRT greater than a fixed cutoff (top 1.6%) are sent for the second tier of testing with a variant panel. The fixed IRT cut-off is changed occasionally to reflect a yearly IRT screen-positive percentile of 1.6%, and the California Department of Public Health evaluates IRT percentiles monthly.[21] In the second step of the CF NBS algorithm, the DBS is then analyzed for a panel of *CFTR* variants only in infants with a high IRT level. The *CFTR* variants included in the panel has changed over time: The initial panel included 28 variants, one variant added in October 2007, nine added in December 2007, and three added in August 2008. This 39 *CFTR* variant panel was used until June 2020, when the panel was expanded to 75 *CFTR* variants (Supporting Information S1: Table 1). Infants with two CFTR variants identified by the panel are reported as a positive NBS. Infants with no *CFTR* variants identified by the panel are reported as a negative NBS for CF. If an infant was found to only have one *CFTR* variant on the panel, *CFTR* sequencing was performed as the third step of the NBS algorithm. Sequencing analysis was conducted using Sanger sequencing at Ambry Genetics until June 30, 2010, and then at Stanford University since July 1, 2010. Infants with two or more *CFTR* variants identified by sequencing are reported as a positive NBS. Infants with zero or one *CFTR* variants identified by sequencing are reported as a negative NBS. The California Department of Public Health encourages the California CF Centers to report people with CF with a false negative NBS back to the California NBS program.

2.2 | Overall design

This is a cohort study of infants with CF born in California from 2007 to 2021 who underwent the CF NBS to examine ethnic differences among those with a false-negative NBS for CF. We analyzed the race and ethnicity of three populations: all infants who had California NBS for CF, all infants diagnosed with CF, and all infants diagnosed with CF who had a false-negative NBS. In the false-negative NBS population, we then examined by race and ethnicity which step of the NBS yielded the false-negative result: low IRT or high IRT with less than two variants detected (with no variants on *CFTR* panel, or no or one variant with *CFTR* sequencing). Infants diagnosed with CF-related metabolic disorder (CRMS) were not included in the analyses.

We compared the detection rate of at least one or two *CFTR* variants in the California CF NBS population born from 2011 to 2021 by race and ethnicity using the original 39 variant panel, the current 75 variant panel, and all the disease-causing variants in the CFTR2

database. In 2022, there were 402 disease-causing *CFTR* variants in the CFTR2 database. Variants of varying clinical consequence in the CFTR2 database were not included in our analyses.

Infant race and ethnicity is reported by the parent at the time of DBS collection and recorded by the phlebotomist. Infant race and ethnicity were defined per 2010 US. Census guidelines as American Indian or Alaska Native; Asian; Black/African American; Hispanic; non-Hispanic White; and other/missing. Hawaiian/Pacific-Islander was combined with Asian due to the small number of infants with CF. The race and ethnicity of infants with multiple races or ethnicities was aggregated to the minimum standard as per the US Department of Health and Human Services Office of Minority Health.

2.3 | Statistical methods

We compared the race and ethnicity of the false-negative NBS population to the entire population screened in California and to the population with CF with a positive NBS. Then, for the false-negative NBS population, we compared each step of the NBS where the false-negative NBS resulted (low IRT, no variants on *CFTR* panel, no or one variant with *CFTR* sequencing) by race and ethnicity. We then examined novel *CFTR* variants identified by race and ethnicity.

Next, we compared racial and ethnic differences in the performance of the 39 *CFTR* variant panel used from 2008 to 2020, the currently used 75 *CFTR* variant panel, and all 402 disease-causing *CFTR* variants in the 2022 CFTR2 database.

All statistical analyses were performed in SAS 9.4 (SAS Institute). Distribution of race and ethnicity by total screened population, total CF population, and NBS false-negative CF population was calculated and differences in proportions were determined with Mantel–Haenszel χ^2 statistics. To measure the association between race and ethnicity and the risk of being an NBS false negative, odds ratios and 95% confidence intervals were calculated using logistic regression for all races and ethnicities with white race as the referent group. Differences in IRT distribution by race/ethnicity was tested by comparing IRT mean values using *T*-tests with white race as the referent group. Case detection rates for three different variant panels were calculated by race and ethnicity by determining the number of cases detected over the total number of cases.

The California Health and Human Services Agency's Committee for the Protection of Human Subjects has determined that the program evaluation and surveillance activities conducted by the California NBS Program represent exempt research per federal guidelines, Section 46.101(b)(4)(ii)). The data provided in this study have been deidentified in compliance with federal HIPAA standards.

3 | RESULTS

In 14 years, 2007–2021, 6.8 million infants in the California NBS program were screened for CF. During those years, 912 infants were diagnosed with CF; 828 were diagnosed after a positive NBS, and 84 infants had a negative NBS but were later diagnosed with CF. Missed

NBS infants with CF represent 0.001% of the total screened population and 9.2% of the known population with CF born in California during 2007–2021.

The race and ethnicity of the infants diagnosed with CF after a false-negative NBS varied from the CF population born in 2007–2021 (Table 1). Among the 84 missed cases, two (2.4%) were American Indian/Alaska Native, 10 (11.9%) were Asian, six (7.1%) were Black/African American, 36 (42.9%) were Hispanic, 29 (35.5%) were non-Hispanic White, and one (1.1%) was other/unknown race and ethnicity. The demographic breakdown of the false-negative NBS cases differed from the total CF population born in California in 2007–2021, as Black/African American (7.1% vs. 4.3%) and Asian (11.9% vs. 3.4%) missed cases were over-represented compared to the overall population while non-Hispanic White (35.5% vs. 47.1%) missed cases were underrepresented. Asian infants with CF (OR 6.3, 95% CI 2.7–14.5) and Black/African American infants with CF (OR 2.5, 95% CI 1.0–6.5) were more likely to have a false-negative NBS compared to non-Hispanic white infants with CF (Table 2).

As shown in Table 2, of the 84 infants with a false-negative NBS for CF, 38 (45.2%) had low IRT levels, and 46 (54.8%) had high IRT values (but no *CFTR* variants on the panel or none or only one additional *CFTR* variants identified by sequencing). Low IRT levels accounted for false-negative NBS results in 100% (2 out of 2) of American Indian/Native Alaska, 50% (3 out of 6) of Black/African American, and 58.6% (17 out of 29) of non-Hispanic White infants (Table 2). Of the 38 false-negative NBS infants with low IRT levels, the diagnosis of CF was made based on meconium ileus in nine infants (24%), a positive family history in 13 infants (34%), and symptoms of CF in 27 infants (71%); the diagnosis in some infants was suspected for multiple reasons. Of these 38 infants, 100% of Asian, Black/African American, and American Indian/Native Alaska infants were diagnosed due to symptoms compared to only 59% of non-Hispanic White infants. In those with a low IRT level, being diagnosed due to meconium ileus was more common in Asian (1 out of 2, 50%), Black/African American (1 out of 3, 33%), and Hispanic (5 out of 14, 36%) infants compared to non-Hispanic White (2 out of 17, 12%) and American Indian/Native Alaska (0 out of 2, 0%) infants. In infants with false-negative NBS due to a low IRT, the majority had IRT values that were far below the cutoff of 1.6%. Adjusting the IRT cutoff to 2% would detect an additional four infants but would result in an additional 21,901 infants being genotyped. Adjusting the IRT cutoff to 3% would detect an additional 10 infants but would result in an additional 89,345 infants being genotyped.

Small variations in IRT levels according to racial and ethnic group (Table 3) are reflected in the percentage of infants in each racial and ethnic group with CF and a high IRT result with <2 variants detected. Out of nearly 6.8 million infants screened in California from 2007 to 2021, the proportion of infants with CF and a high IRT result with <2 variants detected were: American Indian/Native American (1.8%), Asian (1.0%), Black/African American (4.6%), Hispanic (1.5%), other/unknown race (1.8%), and non-Hispanic White infants (1.5%). Having a false-negative NBS result for CF with a high IRT value with <2 variants detected was more likely in Asian (OR 12.2, 95% CI 4.5–32.8) and Hispanic (OR 2.2, 95% CI 1.1–4.5) infants than in non-Hispanic white infants due to not detecting two *CFTR* variants either on panel or by sequencing (Table 2). Asian infants had the

greatest proportion (80%) with <2 variants detected false-negative NBS due to no *CFTR* variant identified on the variant panel (Table 4) among any racial category. Most races and ethnicities had a small percentage of infants with false-negative NBS that had no or only one *CFTR* variant identified on sequencing. Of the 46 false-negative NBS infants with high IRT levels with <2 variants detected, the diagnosis of CF was made based on meconium ileus in 15 (33%), positive family history in 15 (33%), and various symptoms in 34 (74%). Besides no Black/African American infants having meconium ileus in the false-negative NBS with <2 variants detected group, there were little differences between races and ethnicities in how they were diagnosed.

To detect CF in a racially and ethnically diverse population, it is necessary for NBS to include a *CFTR* variant panel that includes the variants of all races and ethnicities. In the CF population born in 2007–2021 in California, we examined the detection of one and two *CFTR* variants by the following variant panels by race and ethnicity: former 39-variant panel, current 75-variant panel, and the 402 disease-causing variants in the CFTR2 database (Table 4). Compared to the 39-variant panel, there was an improvement in the detection of two *CFTR* variants with the 75-variant panel in Black/African American (52.3% ≥ 64.1%), Hispanic (44.8% ≥ 57.2%), and non-Hispanic White infants (61.6% ≥ 72.6%). There was no improvement in the detection of two *CFTR* variants in American Indian/ Native Alaska, Asian, or other/unknown race infants with the 75-variant panel. With the 402 disease-causing variants in the CFTR2 database, there was a large increase in the detection rate of Black/African American (82.1%), Hispanic (70.1%), non-Hispanic White (81.4%), and other/unknown race infants (81.8%). Compared to the 39- and 75-variant panels, there was only a slight increase in the detection rate of Asian infants (28.1% ≥ 31.3%) and no improvement in Native American infants with the CFTR2 database.

As expected, detection of at least one *CFTR* variant was higher than detection of two *CFTR* variants in all races and ethnicities (Table 5). Overall, there was a small improvement in detection rates with at least one *CFTR* variant on the 75-variant and 402-variant panels compared to the 39-variant panel.

Detection of at least one *CFTR* variant was the lowest in Asian infants with all panels; even with all known disease-causing variants, CFTR2 detection was only 80.6%. In all other races and ethnicities, detection was 95% or higher using all panels. With the 402-variant panel, there was 100% detection of at least one *CFTR* variant in Black/African American and other/unknown race infants. There was >98% detection in Hispanic and non-Hispanic White infants.

Overall, 92 variants were detected in the missed cases and four were novel. The majority of variants in infants with false-negative CF screening were only identified in one child. In the false-negative CF screening infants with high IRT levels and <2 variants detected, there were eight variants that occurred in more than one child, but 59 variants occurred in one child only. Similarly, in the false-negative CF screening infants with low IRT levels, 24 of the 30 variants identified were only identified in one child. Fifteen variants were seen in more than one infant with false-negative CF screening and frequencies varied by race and ethnicity (Table 5). There were 10 unknown variants, some of which had been analyzed for large

deletions and duplications and some are still awaiting further analysis for large deletions and duplications.

4 | DISCUSSION

Despite California's robust NBS program for CF with IRT/DNA/DNA sequencing protocol and the addition of additional *CFTR* variants in recent years, some infants ultimately diagnosed with CF have a false-negative NBS result. Overall, among 912 infants born with CF in California, 38 false-negative NBS results (4.2%) were due to a low IRT level, and 46 (5.0%) were due to incomplete detection of *CFTR* variants. The most common step of the NBS where the false-negative result occurred was not the same in all races and ethnicities, although the small sample size for some of the race and ethnicity groups limits the certainty of our findings. These results may be an underestimate as some infants could remain undiagnosed or were later diagnosed outside of California. Missed cases must be minimized as infants with false-negative CF screening results are diagnosed after developing adverse outcomes, including malnutrition, respiratory infections, or lung damage.[22–25]

CFTR variant frequency varies among races and ethnicities based on ancestry and migration patterns, but variants are not homogenous between a race or ethnicity.[17,18,26] Due to *CFTR* variant frequency differences and NBS variant panels being primarily based on European populations, people with CF who are of a race other than non-Hispanic White are more likely to be missed on panels used on NBS algorithms.[16,27] Among the population of infants with false-negative screening results for CF, American Indian/Alaska Native, Asian, or Black/African American infants were over-represented compared to the entire CF population. In contrast, non-Hispanic White infants are vastly underrepresented. While there was no difference in representation between the false-negative NBS population and the entire CF population in Hispanic infants, this was due to intentional improvements to the *CFTR* variant panel with the addition of variants found in the Hispanic population.

Over the history of the California NBS program, 49 *CFTR* variants were added to the second step *CFTR* variant panel to improve the detection of Black/African American and Hispanic infants with CF. When we compared the original 39-variant panel to the current 75-variant panel, one *CFTR* variant was identified in 10 additional infants, and two *CFTR* variants were identified in 20 additional infants. Improvement in the detection of two *CFTR* variants was seen in Black/African American, Hispanic, and non-Hispanic White infants but not in detection among Asian, Native American/Native Alaska, or other/unknown race infants. Using the disease-causing variants listed in the CFTR2 database, one *CFTR* variant was identified in 16 additional infants and two *CFTR* variants were identified in 99 additional infants compared to the 75-variant panel. All groups besides American Indian/Native Alaskan had improvement in the detection of two *CFTR* variants with the 402-variant panel. Detection of one *CFTR* variant improved in Asian, Black/African American, Hispanic, and non-Hispanic White infants. Detection of two *CFTR* variants on a panel likely results in earlier diagnosis and time to being seen compared to only one *CFTR* variant identified due to not requiring the third tier *CFTR* sequencing and certainty of diagnosis by providers.

It is essential for CF NBS protocols to reduce missed cases for infants of all races and ethnicities, particularly Asian and Black/African American. Even with the use of all disease-causing *CFTR* variants in the CFTR2 database, detection of even one *CFTR* variant remained low in Asian infants with CF. NBS protocols with only *CFTR* variant panels have limitations, as infants of minoritized races and ethnicities are more likely to have rare or *de novo CFTR* variants that are not included on panels or in the CFTR2 database.[28] As next-generation sequencing has become quicker and more widely available, the incorporation of sequencing to identify rare variants may improve detection further among infants with a race and ethnicity other than non-Hispanic White. In California, the majority of the *CFTR* variants identified by sequencing in missed cases are unique to a single infant. California NBS is considering using next generation sequencing, which will expand the *CFTR* variant panel list to identify more cases after a high IRT level and speed the unmasking of other unusual or unique variants sequenced by the platform. An expanded variant panel will capture more targeted variants that are CF-causing but will also identify more variants of uncertain significance and novel variants that may not be CF-causing thus increasing the amount of CFTR-Related Metabolic Syndrome (CRMS) called by NBS. More research is needed to identify and characterize *CFTR* variants in Asian and Black/African American people with CF so variants can be added to NBS protocols.

The addition of a very-high IRT level step to NBS protocols is a genetic-neutral approach to improving detection. As this will increase the rate of false positive NBS, the determination of the correct cutoff is essential.[29] As Black/African Americans have higher IRT levels, they will be disproportionately affected by the increase in false-positive NBS results, but they are also currently disproportionately affected by being missed on variant panels. With modeling a very high IRT level of the top 0.1%, we found that it would have detected 55% of the missed cases. However, 75% of those infants would have been detected if the protocol had included a panel of all the known disease-causing variants in CFTR2. The benefit of a very-high IRT level is more significant in CF NBS protocols that screen for a limited number of *CFTR* variants. We found that lowering the IRT level would only slightly improve our detection rate while greatly increasing the number of infants requiring genotyping. This would significantly increase the cost of the NBS program and potentially significantly increase the number of infants identified as carriers or with CRMS.

It is important to note that the missed cases described here are likely an underestimate of the false-negative rate as there are likely children with CF undiagnosed in the community. One of the limitations of our paper is that there are certainly children with CF missed on NBS that have yet to be diagnosed with CF and would not be included in this analysis. Only 90% of infants with CF are estimated to be detected by NBS protocols in the United States.[16,30,31] Racial and ethnic minorities may be less likely to be diagnosed after a false-negative NBS due to the misperception that CF occurs only in the non-Hispanic white population. People with CF from minoritized races and ethnicities have worse outcomes, including higher mortality,[32,33] worse pulmonary disease,[34,35] and increased risk of pulmonary infections.[36–38] Thus, false-negative NBS results and delayed diagnosis likely contribute to health disparities in CF.

NBS programs must be thoughtful of equity in all populations in NBS design to provide early diagnosis for patients from every racial and ethnic background. California's NBS system has been a leader in trying to improve health equity in NBS, with the expansion of the variant panel to include variants seen in racial and ethnically minoritized infants with CF, especially variants seen in Hispanic patients. Despite these efforts, California still had 84 missed CF cases in the 14 years since screening began, which is likely an underestimate. We recommend that NBS programs follow the lead of the California NBS Program and aim to try to detect CF in infants of all races and ethnicities. NBS programs should be aware of IRT differences between races and have a DNA testing protocol that includes variants from all populations served. *CFTR* variant panels should ideally be created to target variants appropriate to the populations in each state. Sequencing is then necessary to uncover rare *CFTR* variants which are not targeted by a panel. There needs to be an easily searchable database so providers can easily determine what CF testing an infant had through an NBS at a given time in any state. The CF Foundation should include guidelines for NBS programs to aim for equity.

All healthcare providers must be aware that the NBS can miss children with CF and remain vigilant. Clinicians should refer to CF centers for sweat testing and clinical evaluation if a patient has symptoms or signs suggestive of CF, despite a negative NBS. There needs to be recognition that people of all races and ethnicities can have CF and racially or ethnically minoritized populations of infants are more likely to have a false-negative NBS result for CF. While NBS for CF has had tremendous success, there is a continuing need to improve the equity of NBS for all infants.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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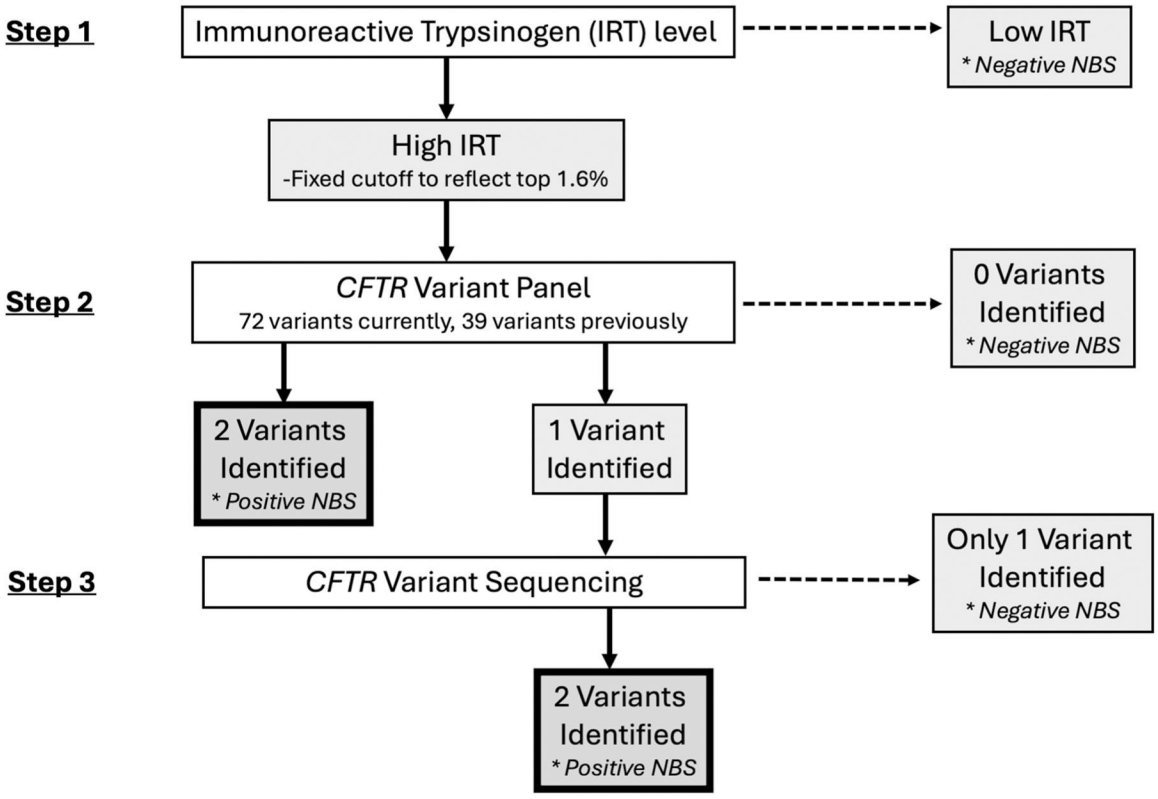

FIGURE 1.
California cystic fibrosis newborn screening algorithm.

TABLE 1

Race and ethnicity of screened population, CF population, and CF population with false-negative NBS (born 2007–2021).

	Screened population	CF population	CF population with false-negative NBS
Total population	6,790,108	912	84
American Indian, Alaska native	65,841 (1.0%)	12 (1.3%)	2 (2.4%)
Asian	969,940 (14.3%)	32 (3.4%)	10 (11.9%)
Black, African American	532,565 (7.8%)	39 (4.3%)	6 (7.1%)
Hispanic	3,326,965 (49.0%)	388 (42.5%)	36 (42.9%)
Other race, unknown	108,775 (1.6%)	11 (1.2%)	1 (1.2%)
Non - Hispanic White	1,786,023 (26.3%)	430 (47.1%)	29 (35.5%)

Note: The overall differences between the distributions in race and ethnicity between populations is: Screened population versus CF population χ^2 $p < 0.0001$; Screened population versus false negative NBS population χ^2 p Value is 0.12; CF population versus false negative NBS population χ^2 p Value is 0.0005. CF population includes all babies with CF born from 2007 to 2021 in California.

Abbreviations: CF, cystic fibrosis; NBS, newborn screening.

TABLE 2

Increased odds for false-negative NBS In Asian and Black infants.

	All false-negative NBS (*n* = 84)				Low IRT false-negative NBS (*n* = 38)				<2 Variants detected false-negative NBS (*n* = 46)			
Race and Ethnicity	***N***	**% missed[a]**	**OR**	**95% CI**	***N***	**% missed[a]**	**OR**	**95% CI**	***N***	**% missed[a]**	**OR**	**95% CI**
American Indian, Native Alaskan	2	17%	2.8	0.6–13.2	2	17%	4.7	1.0–23.2	0	0%	n/a	n/a
Asian	10	31%	6.3	2.7–14.5	2	6%	2.1	0.5–9.9	8	25%	12.2	4.5–32.8
Black, African American	6	15%	2.5	1.0–6.5	3	8%	2.1	0.6–7.7	3	8%	3.0	0.8–11.3
Hispanic	36	9%	1.5	0.9–2.4	14	4%	0.9	0.5–1.9	22	6%	2.2	1.1–4.5
Other, Unknown	1	9%	1.4	0.2–11.2	0	0%	n/a	n/a	1	9%	3.3	0.4–28.2
Non-Hispanic White	29	7%	Ref	-	17	4%	Ref	-	12	3%	Ref	-

Note: The reference group for the odds ratios is non-Hispanic white. The <2 variants detected false-negative NBS group includes infants who either had no CFTR variants identified on the panel or those who had only one or no CFTR variant identified by sequencing.

Abbreviations: CF, cystic fibrosis; IRT, immunoreactive trypsinogen; NBS, newborn screening.

[a]The percentage missed is of the total CF population born in California.

TABLE 3

IRT levels (ng/ML) according to racial/ethnic groups (215,208 infants screened in January-July 2021).

Group	Mean (SD)	Median	*p* Value
All	26.9 (15.4)	23.6	-
American Indian/Native Alaskan	27.2 (15.4)	23.7	0.6
Asian	23.9 (13.3)	20.8	<0.0001
Black/African American	32.4 (18.3)	28.1	<0.0001
Hispanic	26.8 (14.9)	23.7	0.002
Other/Unknown	26.8 (14.7)	23.6	9.4
Non - Hispanic White	27.1 (15.7)	23.9	Reference

Note: The reference group for comparisons in non-Hispanic White.

TABLE 4

Detection of 1 and 2 *CFTR* variants: Comparison of 39 variant, 75 variant, and 402 variant panels for California CF NBS.

	Total number		American Indian, Native Alaskan		Asian		Black, African American		Hispanic		Other, Unknown Race		Non-Hispanic White	
	N	**% Detected**	***N***	**% Detected**	***N***	**% Detected**	***N***	**% Detected**	***N***	**% Detected**	***N***	**% Detected**	***N***	**% Detected**
Total population	912	-	12	-	32	-	39	-	388	-	11	-	430	-
2 Variants detected														
39 Variant panel	480	52.6%	6	50%	9	28.1%	20	52.3%	174	44.8%	6	54.5%	265	61.6%
75 Variant Panel	580	63.6%	6	50%	9	28.1%	25	64.1%	222	57.2%	6	54.5%	312	72.6%
402 Variant panel	679	75.4%	6	50%	10	31.3%	32	82.1%	272	70.1%	9	81.8%	350	81.4%
1 Variant detected														
39 Variant Panel	871	95.5%	11	91.7%	24	75.0%	37	94.9%	367	94.6%	11	100%	421	97.9%
75 Variant Panel	881	96.6%	12	100%	25	78.1%	38	97.4%	372	95.9%	11	100%	423	98.4%
402 Variant Panel	897	98.4%	12	100%	26	81.3%	39	100%	382	98.5%	11	100%	427	99.3%

Note: Case detection rate of 1 and 2 CFTR variants using three different variant panels by race and ethnicity in the CF population born in 2007–2020 in California. The 39-variant panel is the original panel used in the California NBS protocol. The 75-variant panel is the one currently used in the California NBS protocol. The 402-variant panel is all the disease-causing CFTR variants listed in the CFTR2 database in 2022.

Abbreviations: CF, cystic fibrosis; NBS, newborn screening.

TABLE 5

CFTR variants occurring more than once in false-negative NBS cases.

	Variants of all missed cases						
Race and Ethnicity	**American Indian, Alaska Native**	**Asian**	**Black, African American**	**Hispanic**	**Other Race, Unknown**	**Non-Hispanic White**	**Total**
CFTR variant total	4	19	12	66	2	52	155
delF508	1 (25%)	1 (5.3%)	4 (33.3%)	8 (12.1%)	1 (50%)	18 (34.6%)	33 (21.3%)
Unknown	0	1 (5.3%)	0	6 (9.1%)	0	3 (5.8%)	10 (6.5%)
2954delT[a]	0	0	0	6 (9.1%)	0	0	6 (3.9%)
1811 + 1643 G > T[a]	0	0	0	3 (4.5%)	0	0	3 (1.9%)
G542X	0	0	0	2 (3.0%)	0	1 (1.9%)	3 (1.9%)
Q890X	0	0	0	3 (4.5%)	0	0	3 (1.9%)
R117H	0	0	0	0	0	3 (5.8%)	3 (1.9%)
S945L[a]	0	0	0	3 (4.5%)	0	0	3 (1.9%)
3120 + 1 G > A	0	0	1 (8.3%)	1 (1.5%)	0	0	2 (1.3%)
3876delA	0	0	0	2 (3.0%)	0	0	2 (1.3%)
CFTRdele22,23	0	0	0	0	0	2 (3.8%)	2 (1.3%)
L206W	1 (25%)	0	0	0	0	1 (1.9%)	2 (1.3%)
R117C	0	1 (5.3%)	0	0	0	1 (1.9%)	2 (1.3%)
E407X[a]	0	2 (10.5%)	0	0	0	0	2 (1.3%)
delF311	0	0	0	2 (3.0%)	0	0	2 (1.3%)

Note: These are the CFTR variants that occurred more than once in the false-negative NBS cases born in California. Some of the unknown variants include analysis for large deletions and duplications. Some of cases with unknown variants are still awaiting analysis for large deletions and duplications.

Abbreviations: CF, cystic fibrosis; NBS, newborn screening.

[a]These variants were not included in either the 39-variant or 75-variant panels.