Comparing the clinical yield of carrier screening: genotyping versus exon sequencing

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Introduction

Expanded carrier screening (ECS) identifies carriers of recessive diseases and may be performed using either targeted genotyping (TG) or next generation sequencing (NGS). Inspired by recent calls for greater transparency in ECS panel selection[1], here we propose a framework for the systematic design and comparison of ECS panels. Using ECS results from more than 400,000 patients, we use this framework to evaluate the performance of idealized TG and NGS panels.

Systematic design of ECS panels

We propose the following method for ECS panel design:

1. Enumerate severe and profound candidate diseases using systematic severity classification[2]
2. Maximize specificity through careful assay and curation processes
3. Maximize aggregate sensitivity by selecting diseases with high disease risk[3]
4. Maximize the per-gene negative predictive value to yield high confidence in non-carrier status for individual conditions

Previously, carrier frequency (CF) and carrier couple frequency (CCF) have been used to quantify the sensitivity of ECS panels. However, we recently suggested[3] the use of disease risk (DR) for this purpose, where the disease risk is the probability that a random child will be affected by one of the panel diseases.

Method	Carrier frequency	Carrier couple frequency	Disease risk
			
Meaning	400 in 10,000 persons are carriers of this disease	16 in 10,000 couples are carrier couples of this disease	4 in 10,000 children will be affected by this disease
Limitations	Cannot compare autosomal and X-linked diseases	Cannot compare diseases with complex inheritance	Harder to compute

Real-time variant curation allows high sensitivity and specificity

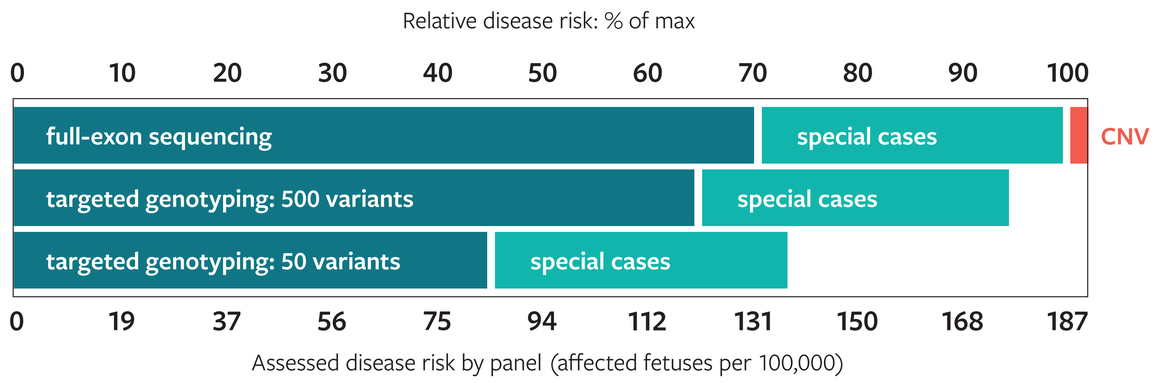
To evaluate variant classification performance, we compared Counsyl *Family Prep Screen* classifications to ClinVar (April 2016 Release)[4]. As a reference standard, we selected 505 variants for which at least two external reference laboratories reached complete consensus. For the sake of comparison, we also considered two fully automated curation approaches based on the Variant Effect Predictor (VEP) algorithm[5]. As compared to VEP, the real-time curations achieve high sensitivity and specificity.

Curation method	Curation type	Gold standard	Sensitivity	Specificity
Counsyl	Real-time	Clinvar Consensus	91.1%	99.7%
VEP Specific	Automated	Clinvar Consensus	34.6%	99.7%
VEP Sensitive	Automated	Clinvar Consensus	95.5%	40.8%

REFERENCES 1. Grody, W. W. et al. ACMG position statement on prenatal/preconception expanded carrier screening. Genet. Med. 15, 482–483 (2013). **2.** Lazarin, GA. et al. Systematic classification of disease severity for evaluation of expanded carrier screening panels. PLoS ONE 2014; 9(12): e114391. **3.** Haque, IS et al. Modeled Fetal Risk of Genetic Diseases Identified by Expanded Carrier Screening. JAMA. 2016;316(7):734-742 **4.** Landrum, M. J. et al. ClinVar: public archive of interpretations of clinically relevant variants. Nucleic Acids Res. 44, D862–8 (2016). **5.** McLaren, W. et al. The Ensembl Variant Effect Predictor. Genome Biol. 17, 122 (2016).

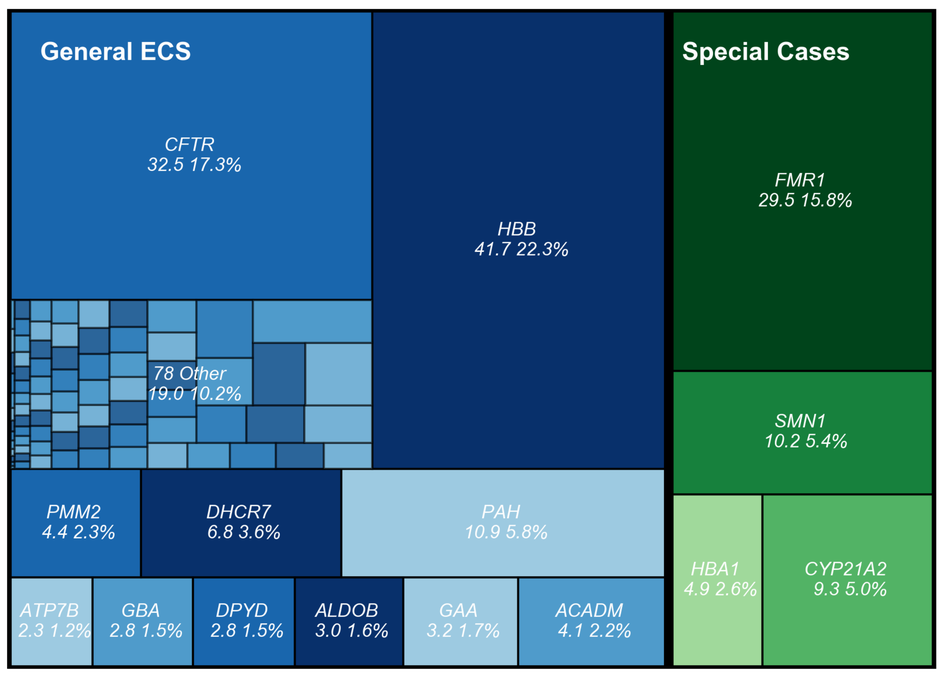
Lessons from idealized panels

To assess the sensitivity of various ECS approaches, we compared the disease risk captured by several idealized panels. We first considered an NGS panel that excludes several "special case" diseases (fragile X syndrome, 21-hydroxylase-deficient congenital adrenal hyperplasia, alpha thalassemia, and spinal muscular atrophy) that are technically challenging to probe. We then considered the effect of adding special cases and panel-wide copy number (CNV) calling. We finally considered "best-possible" TG panels with a fixed number of variants, both with and without the special cases. The disease risk of each idealized panel shows that neglecting special cases and exon-wide coverage overlooks 10% to 55% of affected children.



High-prevalence genes dominate disease risk

A common question is whether an ECS panel would benefit from the addition of more genes. While adding more genes always improves the sensitivity, typically the most prevalent diseases contribute over half of the disease risk. Thus, the way to improve ECS testing may be to improve sensitivity in existing diseases, such as via panel-wide CNV calling, which (as shown in the previous section) contributes approximately 4 affected children per 100,000.



Assessed per-gene disease risk and relative disease risk

Conclusions

Disease risk for severe and profound diseases allows systematic comparison of ECS panel detection power. Idealized ECS panels show that technically challenging genes and full-exon coverage are dominant contributors to overall sensitivity.

Methods

405,195 patients seeking ECS (Counsyl *Family Prep Screen*) between Jan. 2012 and Feb. 2016 for reason of "Carrier Testing" were anonymized and included in the present analysis. TG and NGS based allele counts were combined to reduce statistical uncertainty[3]. Only diseases considered "Severe" or "Profound", as defined previously[2], were included for the present analysis. Results for self-reported ethnicities were reweighted based on US census data.