EDITORIAL

Molecular explanations for variability of clinical phenotypes

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Despite major advances in genomic techniques, multiple challenges still remain in Clinical Genetics—both in identifying causal variants and understanding phenotypic variability. One explanation for the observation that some people with characteristic clinical phenotypes lack detectable genomic variants is mosaicism. In this issue, Francis et al. [1] report a comparison of SNP-microarray technology in saliva compared with blood to detect low-level copy number variant mosaicism in patients with intellectual disability (ID). The study included 23,289 blood samples and 20,857 saliva samples from 6653 individuals with syndromic ID and 37,493 with non-syndromic ID. In 20 participants, a mosaic abnormality was observed in saliva but not in blood. Saliva had a higher diagnostic rate for mosaic copy number variants. One reason for this is when the mosaic cell line is non-haematological.

Chesneau et al. [2] evaluate the role of mosaic genomic variants in eye disease. Microphthalmia, Anophthalmia and Coloboma (MAC) are congenital eye malformations that can be simple or complex, and can be unilateral or bilateral. Despite extensive genomic investigations, most such patients remain without a genomic diagnosis. In 78 such patients, a bioinformatics pipeline designed to detect mosaic variants was applied. No evidence of somatic or gonadal mosaicism was found to explain these conditions.

The lack of causal variants in people with well defined clinical phenotypes can also be explained by the presence of variants that cannot be detected by clinical sequencing or correctly classified. Fortugno [3] reports that Loeys-Dietz syndrome can be associated with truncating variants in the penultimate exon of TGFBR1. These can be detected by exome or genome sequencing but classified as VUS due to lack of functional data. This paper reports functional data confirming the pathogenic nature of these variants and helping solve clinical cases.

Pathogenic large scale inversions of dystrophin, causing Duchenne muscular dystrophy frequently escape detection in clinical testing [4]. Optical mapping identified the pathogenic inversion in DMD locus: inv(X)(p21.1q21.1). Long-read DNA sequencing precisely mapped the breakpoints of the inversion mutation, and the entire 2.2 Mb DMD gene was effectively analysed with a depth coverage of 18.41. The breakpoints of the inversion were located at chrX:32,915,769 and chrX:87,989,329, and the presence of SINE and LTR sequences were found at these locations. The inversion mutation prevented exons 3–55 from being transcribed.

Splice site variants may not be detected by exome sequencing, and if detected may need functional studies to validate them [5]. Ten percent of patients in a large series of individuals with cardiac arrhythmias were found to have splice variants (+/− 10 base pairs from intron boundary). The paper provides useful guidelines on which in silico tools might be best to classify these.

Novel genomics technologies also help our understanding of cancers. Dixon et al use nanopore sequencing to describe structural variation in breast cancer susceptibility genes [6]. Rare variants in genes associated with high-penetrance cancer predisposition syndromes confer a strong genetic susceptibility to breast cancer in 5–10% of cases depending on ascertainment criteria. Nanopore sequencing revealed 14 distinct structural variants in 19 carriers, including three deletions spanning BRCA1 exons 1–2, and a partial BRCA1 pseudogene. Five individuals had deletions of CHEK2 exons 9–10 (which is associated with 1% of breast cancers in Poland). These findings demonstrate the potential for long read sequencing to characterise haplotype-resolved structural variation in personal genomes.

Of course, sometimes lack of phenotype associations with gene variants are due to lack of statistical power in cohorts. Figlioli and colleagues [7] use a large dataset to demonstrate the association of FANCM missense variants and breast cancer risk in a European population. Sometimes of course small populations within larger nations have specific genetic considerations; Kerr et al. identify this with the BRCA1 specific variant in the Orcadian population [8]. Alternatively, we can find that variants we previously thought were restricted to certain populations have broader relevance: as is the case for this ATTR variant in Italy [9]. Sometimes statistical evidence is not enough to classify genomic variants. Torices et al. report functional characterisation of several PTEN variants, which will help with clinical interpretation [10].

The use of genomics in reproductive decision making and reproductive medicine remains highly topical. Freeman and colleagues report broad consensus that hearing loss genes should be included in reproductive carrier screening [11]. The experiences of Dutch women who opted for genome wide non-invasive prenatal testing are also considered in this issue [12]. Wilmot provides a review of antenatal screening practices for Trisomy 21 [13]. Phenotyping remains important in rare disease and this issue presents speech and language assessment in Koolen de Vries syndrome [14].

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ADDITIONAL INFORMATION
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