

Evaluation of the Value of Next-Generation Sequencing in a Series of 66 Fetuses with Complex Cardiac Malformations

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INTRODUCTION: During ultrasound prenatal screening, the discovery of a complex cardiac malformations leads to cytogenetic tests such as FISH caryotype, standard caryotype and/or aCGH. Because of the availability of Next-Generation Sequencing (NGS), we tested whether NGS could yield information on the etiology of some of these cases.

METHODS: After ultrasound discovery of a complex cardiac malformation during pregnancy, a fetal sample was performed after obtaining an informed consent to carry out cytogenetic tests and NGS on a panel of more than 400 genes. DNA was prepared with a customized set of probes (Nimblegen, Roche). Eight DNA samples by run were sequenced on a NextSeq500 (Illumina) with a mid-size flow cell. Sequences were analyzed through a pipeline designed locally (VarAP) able to detect deletion/duplication of exons/gene (DeCovA). Putative causative variants were confirmed by Sanger sequencing and the segregation of selected variants was performed on family members.

RESULTS: 66 fetal DNA were sequenced. A majority of fetuses had heterotaxy and complex cardiac malformations (40 cases) but 26 cases had a severe heart defect (mainly hypoplastic left heart syndrome) but no heterotaxy. A mutation was found in 12 cases (18%) mainly in heterotaxy cases (10 cases) and rarely in non-heterotaxy cases (2 cases). Altogether, 7 genes had a mutation in heterotaxy patients (DNAI1, GDF1 (2x), LMLN2 (2x), MMP21, NEK2, SHH and ZIC3 (2x)) and 2 genes in non-heterotaxy cases (MYH6, TAB2). In most cases, the mutation was recessive autosomal (DNAI1, GDF1, LMLN2, MMP21, NEK2, MYH6) or X-linked (ZIC3) but in 2 cases the mutation was dominant with very variable expressivity (SHH, TAB2). No de novo mutations were discovered.

CONCLUSIONS: Discovering causal gene mutations in fetal cases with complex cardiac malformations is feasible and leads to a specific genetic cause in 25% of fetuses with heterotaxy and less than 8% of cases with no-heterotaxy. Uncovering the genetic cause of fetal cardiac malformation does not change the course of pregnancy which is determined by the cardiac and extracardiac malformations but it allows a genetic counseling to the parents with an accurate evaluation of the recurrence risk for future pregnancies.