Abstract ID:- 112

Abstract Topic:- Molecular and cytogenetic diagnostics

Abstract Title:- Methylation specific polymerase chain reaction to improve diagnostic yield of pediatric disorders with intellectual disability, abnormal growth and behaviour

Presenting author name :- Gayatri Iyer

Presenting author institute:- Kamineni Hospitals, Hyderabad (Previous) and Tata Institute for Genetics and Society (Current)

Co-author name:- Dr. Qurratulain Hasan

Co-author institute:-Kamineni Hospitals, Hyderabad

Aims:-Background: Advances in genomic technologies have helped in the diagnosis of about 7000 genetic disorders. However, more than 25% of the disorders are still undiagnosed emphasizing need for newer modalities. Some mammalian genes are imprinted, i.e they are expressed in a parent-of-origin specific manner. This phenomenon of genomic imprinting, and if altered gives rise to conditions with intellectual disability (ID), abnormal growth and behaviour. IVF pregnancies have a 3-8-fold increased risk for imprinting disorders. Since imprinting is regulated by DNA methylation without altering the underlying sequence, conventional karyotyping or sequencing cannot detect them. In India, though there are several clinical reports of imprinting disorders, very few are molecularly confirmed. The reason for under-diagnosis is lack of awareness in clinicians, absence of referral for appropriate test and lack of laboratories offering methylation-based testing. Objective: 1. To develop simple, cost-effective algorithm in a low resource country like India for diagnosing patients with ID, abnormal growth and behaviour.

2. To demonstrate the rationale and optimal conditions for designing diagnostic MS-PCR assay

Methods:- Patient recruitment using clinical scoring scales, pedigree evaluation, genomic DNA isolation, methylation specific polymerase chain reaction (MS-PCR), karyotyping, next generation sequencing (NGS), fluorescent insitu hybridization (FISH), chromosomal microarray (CMA) with pre and post-test genetic counselling.

Results:- We followed a diagnostic algorithm, which included clinical evaluation and pedigree recording, a cohort of 102 patients were selected for carrying out MS-PCR. 28% of cases were confirmed to have an imprinting disorder ie Angelman Syndrome or Prader Willi, Beckwith Wiedemann or Silver Russel Syndromes based on MS-PCR suggesting that these disorders are not as rare as cited. For some cases, karyotyping (n=3, Ring chromosome 15, translocation Down Syndrome, Mosaic trisomy 17) and NGS testing (n=19 like Bardet Biedel, Rett, Cardiofascioskeletal syndromes and a novel AS like disorder etc) was carried out, which gave an overall diagnostic yield of 46%. FISH helped in determining deletion subtype of AS/PWS and CMA was not helpful in cases negative for MS-PCR.

Conclusions:- MS-PCR is a simple, cost effective, robust test with a short turn-around time and should be offered as the first line of testing in a low resource country for children with ID, abnormal growth and behaviour. The test can be performed in any basic molecular biology laboratory with thermal cycler. Undiagnosed cases should be followed by karyotyping and NGS in a sequential manner to attain good diagnostic yield.

Keywords:- Genomic imprinting, Molecular diagnosis, genetic counselling, rare genetic disorders