Identification and validation of de novo point mutations in patients with intellectual disability

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Intellectual disability (ID), is a common disease that affects more than 1% of human population. Patients with intellectual disability have limited ability to learn and function in daily life. Injury and infection could cause intellectual disability, but most cases are caused by problems in our genetic material. Some of the genetic causes have been found for common syndromes, such as Down syndrome, fragile X syndrome, however, the causes are still unknown for most patients. It is of great interest to identify the cause in undiagnosed patients with ID as it may help in treatment as well as anticipating the development of the disease. Identification of the genetic cause also enables family planning and allows other family members to test for carrier status.

Our genetic information is stored as sequences in our genetic material. When there is a permanent change in a single position in these sequences, we call it a point mutation. When the alteration in the genetic material is present for the first time in one family member as a result of a point mutation in a germ cell (egg or sperm) of one of the parents or in the fertilized egg itself, it is called a de novo point mutation. One of the possible genetic causes of intellectual disability is de novo point mutations.

The sequences in our genetic material can be categorized as coding regions or non-coding regions. Coding regions is a small part of our genetic material, but contain important information on all components of our body. Non-coding region is much larger than coding region, but their functions are less known. The sum of all the coding regions is called the exome. Mutations in the exome are much more likely to cause disease compared to mutations in non-coding regions. Therefore capturing and sequencing of the exome is a viable and inexpensive strategy.

De novo point mutations have been hard to identify, until we have the technique to massively sequence our genetic material. In this study, we used an exome capture technique and the massively sequencing technique to find causative de novo point mutation in a family trio, which includes one affected child and two healthy parents.

The genetic materials from all three family members were extracted from blood samples. Exomes were captured from the genetic materials. Massively sequencing technique generated the exome sequences of all three family members. These sequences were analyzed to identify possible causative de novo point mutations. This was done by comparing the patients' sequences with the human reference sequences and the parents' sequences, identifying mutations, and filtering out the mutations that were not causative de novo point mutations. A total of 13 candidate mutations were identified and validated by Polymerase Chain Reaction (PCR) and Sanger sequencing.

PCR is a technique to amplify genetic material from low amount to huge amount, using biological machinery. Sanger sequencing is an alternative sequencing technique which has a lower output but higher accuracy than the massively sequencing technique. None of the 13 candidates could be validated by Sanger sequencing in this study. However, the method we used has shown to be effective, and will be more effective with fast developing technologies.

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