Developmental delay referrals and the roles of Fragile X testing and molecular karyotyping: A New Zealand perspective

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Global developmental delay (GDD) affects ~1-3% of children, many of whom will also have intellectual disability (ID).

Fragile X is the major genetic cause of GDD with intellectual disability in males, accounting for ~20% of all X-linked MR.

Majority of developmental delay (DD) cases referred to our laboratory are concerned with the exclusion of a diagnosis of Fragile X, along with simultaneous karyotype analysis to confirm chromosome aberrations.
Global developmental delay is defined as delay in 2 or more areas such as gross motor, fine motor, language, social, cognitive function.

There is no specific criteria for NZ but a paediatrician or other child development specialist makes the diagnosis if they feel that a child has delay in more than one area i.e. if a child was just slow to walk they would be considered to have gross motor delay but if that same child was slow to walk and talk then they would have global developmental delay.
Chromosomal aberrations are the most common cause of GDD/ID.

Historically, the majority of cases referred to a diagnostic laboratory have been analysed using G-banded karyotyping, which detects microscopic genomic abnormalities >5-10 Mb.

Fragile X testing is commonly used to ‘rule out’ Fragile X expansions because of its strong association with X-linked intellectual disability.
but times are changing......

- Implementing array-based technology in place of traditional G-banded karyotyping has increased the frequency of diagnosis among individuals with DD.

- A report documented that in patients with GDD and/or ID, array testing (also known as molecular karyotyping) is diagnostic in ~8% of cases, with G-banded karyotyping accounting for ~4%.

- Furthermore, molecular laboratories have experienced a falling positive detection rate. UK data revealed a 3% positive frequency in 1993 (with only 50% of tests being for children <6 years) compared with a 0.6% positive frequency in 2008.
UKGTN criteria for Fragile X testing

- The UK Genetic Testing Network (UKGTN; http://www.ukgtn.nhs.uk/gtn/Home) has reported that 17% of all tests requested of a molecular laboratory are for Fragile X

- The UKGTN has implemented specific testing criteria for male and female children to increase the testing efficiency of Fragile X in UK laboratories
Currently, our own laboratory is experiencing referrals for simultaneous molecular karyotyping and Fragile X testing.

With increasing clinical laboratory workloads we thought it prudent to review our detection frequencies for both Fragile X and molecular karyotyping.

In light of the falling detection frequencies for the former experienced in the UK, but with an increase in detection frequencies for the latter.
Study to review detection rates, FX and molecular karyotype

- 2046 peripheral blood samples referred for fragile X over 4 years

- During this 4 year period we also implemented molecular karyotype (MKAR) analysis in place of G-banded karyotype for all DD referrals

- 443 developmental delay cases have been tested for both Fragile-X and molecular karyotype.
Results-Fragile X

- 2046 peripheral blood samples referred for fragile X over 4 years

- 13 were positive for a full mutation with no known family history. 3 ♀ and 10 ♂

Mutation detection frequency for Fragile-X of 0.6% with no known family history.
Results-MKAR

443 cases referred for DD tested for both Fragile X expansion and MKAR

We have detected clinically significant imbalances in 26 cases: a detection frequency of 5.9%.

In addition to the clinically significant imbalances, we detected 66 cases with imbalances of uncertain significance and 11 cases with long contiguous stretches of homozygosity, which accounts for a further 17% of all patients (data not shown)
Conclusions

• Our data reveal a mutation detection frequency comparable with the UK detection levels of 0.6%.

• Concentration of assessment and community paediatric input to detect delay earlier in children has led to a shift to younger age in the target population for testing for which there is no specific younger age limit.

• Range of problems in children presenting for assessment at this age is much wider, and hence a focus on the level of delay appropriate for Fragile X is less likely.

• Characteristic physical features reported for adults are less applicable at this younger age.

• Parents of younger children are less likely to have completed their families, so there is more urgency for diagnosing or excluding Fragile X.

• The expertise of clinicians for recognising behavioural/personality pointers to fragile X in young children may be less widespread.
Conclusions

- Importantly, there is a 9-fold greater chance of detecting a chromosomal aberration of clinical significance using a molecular karyotype approach than of detecting a full CGG repeat expansion in the FMR1 gene in cases with DD [24 of 443 cases (5.4%)].

- This mutation detection variance may be higher as we have excluded long contiguous stretches of homozygosity (suggestive of autosomal recessive mutations) and results of unknown clinical significance, which account for a further 17% of all DD referrals.
Bigger Conclusions

• Based on this data, Fragile X testing is an inappropriate first-tier test for DD referrals with no known family history of Fragile X.

• Secondly, a greater diagnostic yield is provided by a molecular karyotype.

For 2014 approx. 30% of our molgen tests were for FX!!
Proposed strategy

Specialist referral

Molecular karyotype

Positive result

Final Report

Negative result

Fragile X testing

Actioned upon confirmation from specialist referrer
Future strategy

Specialist referral

Molecular karyotype

Positive result
- Final Report

Negative result
- Whole exome testing
  - Positive result
    - Final Report
  - Negative result
    - Fragile X