Introduction to Molecular Newborn Screening EQAs

- Newborn screening is performed within the first 8 weeks of life to screen for diseases which are treatable if detected early.
- These disorders include Medium chain acyl-CoA dehydrogenase deficiency (MCADD) and cystic fibrosis (CF).
- Medical laboratories working to International Standard ISO15189 are required to participate in these External quality assessments (EQAs) if they provide the molecular screening.
- EQAs for CF and MCADD testing on bloodspots were set up in 2008 and 2010 respectively.
- Samples are distributed four times per year, with a total of 12 samples annually.
- Laboratories are required to test the bloodspot cards using their standard testing method and report genotyping results only.
- Laboratories can score a maximum of 2.0 marks for the genotyping. Deductions are made for incorrect genotyping and nomenclature errors (based on HGVS guidelines).

GenQA Bloodspot Samples 2008-2019

- Since 2008, 116 CF EQA samples have been distributed for this EQA. The samples include the 4 most common pathogenic variants: p.Phe508del, c.489+1G>T, p.Gly551Asp and p.Gly552Ter, as well as “No CFTR variant”, one single rare mutation and compound heterozygotes. The majority of compound heterozygotes included the pathogenic variant p.Phe508del and another rarer variant. A summary of the samples sent is shown in figure 1.
- Since 2010, 96 MCADD EQA samples have been distributed for this EQA. This EQA is only testing for the presence or absence of the most common MCADD variant: c.985A>G. A summary of the samples sent is shown in table 1.

<table>
<thead>
<tr>
<th>Table 1: Summary of samples provided for MCADD EQA</th>
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<tbody>
<tr>
<td>Validated result</td>
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<tr>
<td>c.985A&gt;G absent</td>
</tr>
<tr>
<td>c.985A&gt;G</td>
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<tr>
<td>c.985A&gt;G / c.985A&gt;G</td>
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Participation

- The number of participants in the CF EQA has increased by over 50% since the EQA was first established in 2008. The greatest increase in participation is seen in non-UK laboratories with the number increasing from 3 in 2008 to 10 in 2017-18.
- A summary of the participation numbers for the CF EQA is shown in figure 2.
- The number of laboratories participating in MCADD EQA has remained consistent since the EQA was established, however the number of UK laboratories has decreased from 10 to 7 since 2010.
- A summary of the participation numbers for the CF EQA is shown in figure 3.

Common Errors and Changes in the EQAs

- Throughout the years there has been a very high standard of results submitted for these EQAs. In total for the CF EQA 16 genotyping errors have been reported and 1 genotyping error for the MCADD EQA.
- In two cases, the genotyping errors in the CF EQA were due to apparent sample swaps within the receiving laboratory which resulted in these laboratories incurring 2 genotyping errors each.
- The majority of small errors resulting in arising in these EQAs were due to minor errors in the use of nomenclature. This varied from using out of date nomenclature, incorrect use of brackets and only providing the DNA level or protein level nomenclature.
- When samples were sent which included a stop codon, this was displayed in several ways: "Ter", "*" and "X".
- When there were changes in HGVS guidelines, laboratories were able to use the old and new guidelines when reporting results during the overlap without incurring any deductions to marking.
- A summary of the errors that have occurred in these EQAs is shown in table 2.
- An interesting common occurrence in the CF EQA, was when a single rare mutation was sent out, a large number of laboratories did not detect this. This is due to the fact that many UK laboratories initially only test for the four most common UK CFTR variants and only if one of these is detected will further testing be done. Therefore the rare variant was not detected.
- Some examples of the current HGVS guidelines for some CF and MCADD variants can be found in table 3.

Summary

- Since the CF bloodspots EQA was established in 2008, there has been a total of 116 samples distributed which included 24 different genotypes and has had participation from 14 different countries.
- Since the MCADD bloodspot EQA was established in 2010, there has been a total of 96 samples distributed to 5 different countries testing for the common MCADD variant: c.985A>G.
- Within these EQAs there is a very high standard of reporting, with the majority of errors occurring due to minor mistakes in nomenclature.

For further information or to take part in the next bloodspot EQAs visit us at STANDARD #672 or email info@genqua.org

Acknowledgements

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