Clinical Utility of Whole Exome Sequencing in the Diagnosis of a Child

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The following relationship(s) exist related to this presentation: No relationships to disclose.
Exome/Genome Sequencing

• To date has focused on:
  – Celebrity individuals
  – Familial disease
  – Collections of individuals with a well defined syndrome

• For true clinical utility the technology must be applicable to a simplex case
Clinical Case

• Male who presented at 15 months with poor weight gain and a perianal abscess.
• Symptoms progressed over a few months to an aggressive, refractory inflammatory bowel disease.
• Pathological studies revealed focal granulation tissue with chronic active granulomatous inflammation, consistent with a severe Crohn’s disease.
Clinical Course

• In spite of aggressive medical and immunomodulatory therapy the disease progressed with:
  – mucosal inflammation
  – strictures
  – enterocutaneous fistulæ
  – poor cutaneous wound healing

• ultimately requiring a total colectomy.
Immunological work-up

• anti-neutrophil antibody
• abnormal chemotaxis of neutrophils
• decreased NK cytotoxicity, but no evidence of HLH
• memory skewing of CD4 cells
• inverted CD4 to CD8 ratio
Diagnosis?

• Several forms of immune dysfunction have been associated with inflammatory bowel disease
• May respond to immune reconstitution or require alternate treatment plan dependent on the underlying cause
• Immune reconstitution is a risky procedure
3 options

• Continue current treatment
• Blindly attempt alternate risky therapies
• See if we could obtain information to make a more informed choice
## Variant analysis summary

<table>
<thead>
<tr>
<th>Category</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>High Confidence variants</td>
<td>16,124</td>
</tr>
<tr>
<td>Genic variants</td>
<td>16,012</td>
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<tr>
<td>Protein coding</td>
<td>15,272</td>
</tr>
<tr>
<td>Non-synonymous</td>
<td>7,157</td>
</tr>
<tr>
<td>Two variants in gene predicted to be damaging</td>
<td>67</td>
</tr>
<tr>
<td>- Altering highly conserved amino acids</td>
<td>19</td>
</tr>
<tr>
<td>- Not known to contain many missense mutations</td>
<td>5</td>
</tr>
<tr>
<td>Novel (dbSNP129) Non-synonymous</td>
<td>878</td>
</tr>
<tr>
<td>Homozygous or hemizygous</td>
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<tr>
<td>- Predicted to be damaging</td>
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</tr>
<tr>
<td>- Novel (against dbSNP 130)</td>
<td>8</td>
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<tr>
<td>- Altering highly conserved amino acid</td>
<td>4</td>
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<tr>
<td>- Not found in reference genome sequences</td>
<td>2</td>
</tr>
<tr>
<td>- Not known to contain many missense mutations</td>
<td>1</td>
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</tbody>
</table>
Sanger validation

<table>
<thead>
<tr>
<th>Normal reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>205 G</td>
</tr>
<tr>
<td>310 T G</td>
</tr>
<tr>
<td>315 G A A</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Child</th>
</tr>
</thead>
<tbody>
<tr>
<td>100 T G G</td>
</tr>
<tr>
<td>105 G A A</td>
</tr>
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</table>

The child is hemizygous (single X chromosome copy in a male) for the variant.
Follow-Up

• Mutation independently confirmed
• Because of risk of lymphoproliferative disorder decision made to perform BMT
• Almost total remission of bowel disease following immune reconstitution
• We demonstrated that Genomic sequencing is a useful advance in DNA diagnostic testing that can inform clinical decision making.
Others could benefit

- After initial success significant interest in applying technology to other cases
Ethical Concern

• When sequencing a whole genome, not all disease associated variants will be pertinent to the question at hand

• Although, this is a problem occasionally encountered with clinical array CGH it is expected that this will be seen with increased frequency when performing WGS
• Resources to analyze data and obtain consent are limited
Case Selection- Principals

• Equity of access

• Reserved for individuals in whom
  – the likelihood of success is high
  – reasonable clinical testing has not achieved answer
  – molecular diagnosis has the potential to advance clinical decision making
Two Step Process

• Phase 1 - Nomination
• Phase 2 – Review group
Phase 1 - Nomination

• Two physicians with expertise in disease area determine:
  – Standard clinical assessments have been utilized
  – WGS is clinically warranted in the context of management of the patient and their family
  – Patient/Family is interested in considering WGS

• Referred to:
  – Genetics to initiate consent process
  – Review group
Review group - Members

- Chair – Hospitals Chief Medical Officer
- Three physicians with expertise in the area of interest not directly involved in the case
- Chair of hospital ethics committee
- Medical College ethicist
- Geneticist
- Genomics expert
- Genetic counselor
Review group - Process

• Nominating Physician presents case
• Review group determine:
  – What disease information is related to the clinical question
  – Ensure appropriate clinical consent is obtained
  – Ensure appropriate research protocol and consent are in place if information will be used for research
Confirmatory testing

• We require all DNA testing in our laboratory to be confirmed on a second extraction, preferably by a second technique
• WGS not considered definitive/medically actionable until confirmed
Consent for Data return - Pediatrics

• Ethical opinion:

• “the return of any or all of this [genomic] information was morally permissible and that such a decision as to what should be returned should remain at the discretion of informed parental choice.”
Categorical model for choice

• Decided that parents should preemptively be asked what data they would like returned.
• This is part of the consent process
• Consent by Genetic Counselor and one of two named Clinical Geneticists
• Typical face to face time 6-9 hours with additional telephone calls
Categories
Information actionable in the child in childhood

• “if such results are discovered there is a duty of care to confirm and act upon these results. There is no opt out if such results are discovered”

• There is no obligation on the physicians/testing Lab to actively analyze the data to discover such abnormalities as this is not the focus of the test.
actionable disease with adult onset

• Examples would include BRCA1/2, Hypercholesterolemia
non-actionable disease with onset in adulthood

- Examples: Parkinson, Huntington’s