Background

- Public databases of clinically observed variants are a rapidly growing and valuable resource for laboratories, clinicians, and patients.1,2
- However, variant classification differences between public databases have been raised as a concern by at least one commercial laboratory who suggested that “widespread disagreement” in these databases should “preclude their wider use in clinical practice.”3
- The clinical impact of these disagreements is not clear. Experienced lab directors never simply copy classifications from any public database. Instead, they critically evaluate evidence and determine classifications rigorously following established guidelines.
- Appropriate practices for the use of public databases have been established in the clinical genetics community for years. The need for, and methods for, the integration and quality control of databases by their users are well-understood.
- Our prior studies show that expert BRCA1/2 variant classifications, appropriately utilizing public data (including databases and the literature), are highly concordant with classifications that utilize non-public, proprietary information.
- Here we sought to measure BRCA1/2 classification concordance in a large multi-laboratory public data set.

Prior Studies

- Our recent study4 observed high (99.8%) concordance of 975 BRCA1/2 reports classified following current guidelines using only publicly available data, compared to tests that also utilized non-public information. The study was a blinded analysis in a prospectively accrued, clinically representative patient population (for details see the Methods section of reference 4).
- Our companion clinical utility study5 incorporated additional data in which no classification differences were observed. VUS (variant of uncertain significance) rates were comparable.

II. Variant Classification Process

Variants were classified at Invitae using a system (called Sherlock6,7) that closely adheres to the 2015 ACMG guidelines8 for the Interpretation of Sequence Variants.

Classification Concordance Per-Variant

- Only 27 out of 1800 variants with any discordant classifications between any two labs were observed.
- Counting each variant separately, concordance between pairs of labs is high: 97.2% to 100.0%.
- All of the discordant classifications were in rare variants that, by definition, are present in very few patients.
- Thus, this calculation greatly underestimates the much higher concordance observed on a per-patient basis.
- Reports from other labs that pre-date SCRP releases had comparable concordance to those that post-date SCRP, suggesting that the SCRP data did not bias the other labs.

Methods

I. Data Sources

ClinVar

Approx. 20,000 Patients Represented

Table 2. Classification concordance between laboratories on a per-variant basis.

Table 3. Data used to compare classifications

<table>
<thead>
<tr>
<th>Source</th>
<th>Total Variants</th>
<th>Reported by Multiple Labs*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ambry Genetics</td>
<td>2793</td>
<td>1502</td>
</tr>
<tr>
<td>Myriad Genetics via SCRP</td>
<td>2067</td>
<td>1184</td>
</tr>
<tr>
<td>Invitae</td>
<td>1479</td>
<td>1082</td>
</tr>
<tr>
<td>GeneDx</td>
<td>1214</td>
<td>937</td>
</tr>
<tr>
<td>Counsyl</td>
<td>272</td>
<td>256</td>
</tr>
<tr>
<td>CHEO Molecular Genetics Lab</td>
<td>257</td>
<td>216</td>
</tr>
<tr>
<td>Emory Genetics</td>
<td>203</td>
<td>183</td>
</tr>
</tbody>
</table>

SCRP = Sharing Clinical Reports Project (at UCSC). Most SCRP reports are from 2011 or later. The older Myriad BIC data were not used.
CHEO = Children’s Hospital of Eastern Ontario

* i.e., the number of variants classified by two or more labs on this list.

II. Comparison Basis

Table 4. Comparisons in this study distinguish between potentially clinically actionable and not actionable findings.

<table>
<thead>
<tr>
<th>Positive</th>
<th>Uncertain</th>
<th>Negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>✔</td>
<td></td>
</tr>
<tr>
<td>Uncertain</td>
<td>✗</td>
<td>✗</td>
</tr>
<tr>
<td>Negative</td>
<td>✗</td>
<td>✔</td>
</tr>
</tbody>
</table>

Conclusions

- Classification concordance needs to be measured carefully in order to avoid over-counting differences and misinterpreting the implications for patient care.
- What matters most is the fraction of patients, not the fraction of variants that show a classification discordance that would significantly change management.
- While such disorders are infrequent, they are important and it is essential to resolve them collaboratively, not competitively, in order to deliver the best patient care as in other areas of medicine.1,2
- Laboratories who do not contribute to public databases may be arbitrary and unverifiable claims regarding variant classification.
- Independent review of classifications, such as this study, are enabled by public databases. Such analyses both enhance laboratory quality control and help improve clinical guidelines.
- Even after detailed re-examination of all evidence underlying the few classification disagreements seen in this study, the maximally correct classification under current ACMG guidelines was still unclear. Most classification differences appear to be due to a difference in precise criteria used, not a difference in underlying data available to the lab.