Finding Patients Through Genomic Sequencing

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Disclosures and Disclaimers

I am an employee of and hold equity in Illumina.
Rare Disease
In the NICU, birth defects or genetic conditions attribute to 2–6

~50% of rare diseases impact children

~30% of all admissions

Up to 40% of all deaths

The Diagnostic Journey is Long, Costly and Imposes Significant Burden on the Healthcare System

Identifying all the known rare and ultrarare diseases can remain a challenge even for the most experienced clinical specialists.

RareX: Be Counted

**Purpose**

- Determine more accurate count of RD that resonates with communities
- Offer recommendations for RD communities

**Findings**

- RD Count
  - Conservative count of 10,867 RDs
    - 6,282 Orphanet + 2,065 OMIM + 2,520 OMIM with narrow match to Orphanet RD
  - 87% have a genetic or suspected genetic
  - Treatment options available for ~5%

https://rare-x.org/wp-content/uploads/2022/05/be-counted-052722-WEB.pdf
Comparison of WGS to Standard Testing
Comparison of Genetic Testing

- **Single Gene Testing**: Testing a single gene of interest
- **Targeted Sequencing**: Testing a subset of genes related to a particular indication
- **Whole-Exome Sequencing**: Testing the full exome
- **Whole-Genome Sequencing**: Testing the full genome
### WGS Provides the Most Comprehensive Analysis of Genomic Variants Among All Clinical Genomic Testing Methods

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<tr>
<th></th>
<th>Sanger*</th>
<th>Targeted NGS*</th>
<th>PCR*</th>
<th>CMA*</th>
<th>WES*</th>
<th>WGS*</th>
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<tr>
<td>SNVs</td>
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<td>Structural variants</td>
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*Variant detection may vary depending on laboratory and test offering.

CMA=chromosomal microarray; CNV=copy number variant; FISH=fluorescence in situ hybridization; Indel=small insertion/deletion; NGS=next-generation sequencing; PCR=polymerase chain reaction; SNV=single nucleotide variant; WES=whole-exome sequencing; WGS=whole-genome sequencing.

References:
The Value of Comprehensive Testing
Nearly 10% of molecularly diagnosed patients have multiple pathogenic variants

Significant diversity in pathogenic variation underlying genetic disease.

Multiple variant types identified in 5–12% of molecularly diagnosed cases.1-3

Demonstrates value of comprehensive approach.

Patient variant distribution
(N=699 patients)3

3. Data on file at Illumina Clinical Services Laboratory; patient cohort of 699.
Comparison of WGS to Standard Testing

Rapid End to the Diagnostic Odyssey

MCA: Multiple congenital anomalies.
DD / ID: Development delay/intellectual disability.
FISH: Fluorescence in situ hybridization

Comparison of WGS to Standard Testing

Rapid End to the Diagnostic Odyssey

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Suggestive of a SPECIFIC genetic condition

Phenotypic Presentation

Confirmatory Assay

No

Yes

Genetic disorder suspected

DIAGNOSIS?

NONSPECIFIC (Dysmorphism, MCA, DD / ID)

Targeted assays
- Single Gene Sequencing
- Multi Gene Sequencing
- Mitochondrial DNA Testing
- FISH
- Repeat Expansion (PCR)

Genome-wide assays
- Karyotype
- Chromosomal Microarray (CMA)
- Whole Exome Sequencing (WES)
- Whole Genome Sequencing

Whole-Genome Sequencing
Multiomics publications on the rise—Since 2012, there has been a 63% average year-over-year increase in the number of publications featuring multiomic data.¹

Grant funding growth for multiomics—Since 2012, there has been a 48% average year-over-year increase in the number of active or starting grants for multiomic studies.¹
Clinical Utility of WGS

Utility Is Not One Thing

Clinical utility
- Impact of a result on medical decision-making
- Comparison to other diagnostic tests
- Availability of effective interventions
- Avoidance of invasive testing and ineffective interventions
- Outcomes
- Physician behavior — medical error reduction

Personal utility
- Effect on future reproductive decisions
- Impact on prognosis and direction of care
- Impact on educational and rehabilitative services
- Cascade testing of relatives
- Satisfaction

Economic utility
- Costs of testing
- Cost effectiveness
- Utilization
- Risk reduction

System utility
- Services planning
- Resource allocation
- Access to care
- Efficiency of utilization
- Risk management
- Surveillance

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Supportive Evidence
Clinical Utility of WGS

Rapid-WGS Has a Higher Diagnostic Yield Enabling Impact on Clinical Management of NICU / PICU Patients

Dx yield from head-to-head comparison studies

- Willig (2015) n=35
  - WGS: 57%
  - Standard Tests: 9%

- Petrikin (2018) n=32
  - WGS: 31%
  - Standard Tests: 3%

- Famaes (2018) n=42
  - WGS: 43%
  - Standard Tests: 10%

- Wu (2021) n=202
  - WGS: 37%
  - Standard Tests: 20%

- Kranz (2021) n=354
  - WGS: 31%
  - Standard Tests: 15%

Dx yield from all other peer-reviewed publications

- Soden (2014) n=15
  - WGS: 73%

- Mestek-Boukhibar (2018) n=24
  - WGS: 42%

- French (2019) n=195
  - WGS: 21%

- Kingsmore (2019)* n=118
  - WGS: 25%

- Sanford (2019) n=38
  - WGS: 45%

- Wang (2020) n=130
  - WGS: 48%

31-57% vs. 9-15%

A change in clinical management was reported across 8 of these studies.²⁻⁵,⁸⁻¹¹

Note: Standard Tests may include CMA, FISH, karyotype, targeted gene panels, microarrays, methylation, or other. In Kranz (2021), the comparison was between WGS early arm (15 days after enrollment) vs WGS delayed arm (60 days after enrollment). Cross-trial comparisons cannot be made given different study parameters/design. WGS: whole-genome sequencing

References in slide notes; * Includes both rapid WGS and ultra rapid WGS.
Compared with Standard Approaches, WGS Has Higher Diagnostic Yields in Pediatric Outpatients

Dx yield from head-to-head comparison studies

- Lionel (2017) n=103: 41% vs. 24%
- *Lionel (2017) n=70: 50% vs. 37%
- Costain (2018) / Stavapoulos (2016) n=100: 13% vs. 41%
- Lindstrand (2019) n=100: 27% vs. 12%
- Vanderver (2020) n=34: 59% vs. 16%

27-59% vs. 12-37%

Dx yield from all other peer-reviewed publications

- Gilissen (2014) n=50: 42% vs. 21%
- Taylor (2015) n=156: 36% vs. 24%
- Bick (2017) n=22: 19% vs. 24%
- Thiffault (2018) n=80: 68% vs. 40%
- ***Splinter (2018) n=165: 68% vs. 40%
- Riley (2020) n=40: 33% vs. 24%
- **Turro (2020) n=660: 68% vs. 40%
- Bertoli-Avella (2021) n=534: 19% vs. 24%
- Stranneheim (2021) n=3219: 68% vs. 40%
- Scocchia (2019) n=60: 20% vs. 24%
- ****Bertoli-Avella (2021) n=473: 68% vs. 40%

A change in clinical management was reported in across 7 of these studies.3,5,7,8,11,14,16

Note: Standard Tests may include CMA, FISH, karyotype, targeted gene panels, microarrays, methylation, or other. Cross-trial comparisons cannot be made given different study parameters / design

WGS: whole-genome sequencing

References in slide notes; *Subset of 70 patients with WGS and WES; **(Turro 2020) Neurodevelopmental disorders cohort; ***Note: in subset with negative WES, WGS diagnostic yield was 16%; **** (Bertoli-Avella 2021) 83% of the sample at the age of testing are pediatric population (<16 years of age)
Whole-Genome Sequencing Demonstrates High Diagnostic Yield in Certain Neurological Indications

1. Studies may include combined adult and pediatric cohorts.
*Developmental delay/intellectual disability; ^Epilepsy; # Other neurologic disorders (e.g., leukodystrophies, neuromuscular, movement).

18
Cost-effectiveness of ES/GS for children with rare disease
Modeled estimates of ES and GS compared to standard of care

Key Takeaway
First-line GS is cost-effective (CE) for diagnosing rare disease in infants. GS may be cost-effective in all children under certain assumptions. Standard of care testing prior to ES/GS increases cost without improving outcomes.

Method
- CE model incorporating costs, dx yield and quality-adjusted life years based on publicly available data and published evidence.
- Comparison of 7 test strategies over 10-years and lifetime (ie. 3 scenarios based on assumptions of disability/quality of life.
- Two cohorts of children with suspected genetic conditions analyzed: critically ill infants (<1 year) and all children (<18 years).

Findings
- Standard of care (SoC) testing had lowest cost over 10 years, but also the lowest dx yield.
- **First-line GS projected to increase costs & had higher dx yield** (~$15,048/added dx for infants and $27,349/added dx for all children).
- **First-line GS was most CE** (the lowest additional cost for each additional diagnosis) of the 6 strategies compared to SoC.
- Per probabilistic sensitivity analysis, first-line GS was the most CE approach at commonly accepted willingness to pay thresholds (how much society is willing to pay for a given outcome).
- **Reanalysis of ES or GS in undiagnosed children was considered CE** (good value for money)

Farnaes reported that rWGS reduced length of hospital stay by 124 days and resulted in a net savings of $804,200
• N=42 critically-ill infants in NICU

Dimmock reported that the GS testing in studied cohort cost $1.7 million, but led to $2.2–2.9 million cost savings in hospital costs and professional fees
• N=184 critically-ill infants

Lavelle reported, first-line GS may be the most cost-effective strategy for infants with rare conditions
• WGS is more cost-effective than WES in severally ill infants
• If used first WGS is cost-saving in severely ill infants compared to other common genetic testing in this population; CMA, WES, WES+CMA

Published September 5, 2022, Lindstrand et al, Genetics in Medicine

Our findings strongly suggest that genome analysis outperforms other testing strategies and should replace traditional CMA and FMR1 analysis as a first-line genetic test in individuals with ID/NDD. GS is a sensitive, time- and cost-effective method that results in a confirmed molecular diagnosis in 35% of all referred patients.
Growing Coverage in the US and Globally

**US**

- As of December 2020, commercial insurance plans in the US representing >16M lives have positive medical policies for rapid-WGS in the NICU/PICU
  - Blue Shield of CA; Priority Health, Florida Blue, Horizon, Blue Cross Blue Shield Federal Employees Campaign, Capital Health Plan, Blue Cross of Idaho

- 11 state Medicaid programs have payment rates posed for WGS

**Europe, Middle East & Africa**

- In the UK, **NHS England covers WES and WGS** in clinical routine for defined rare diseases and cancers. **Wales** has also commissioned the use of WGS in critically-ill children.

- Since 2021, **Germany reimburses WGS/WES** with no prior authorization requirement at the national level. In addition, major German health insurance companies also cover WES and WGS at a higher reimbursement level to allow for testing of patients and their parents (‘trios’).

- Other countries covering WES and WGS for patients with undiagnosed, suspected genetic diseases include **Switzerland, Denmark and Sweden**.

- Several countries, including but not limited to **Belgium, France, the Netherlands, Israel and Spain** are actively pursuing integration of WGS into clinical care and/or launching genomic initiatives that would include WGS for rare and undiagnosed disease

**Asia**

- In Nov 2019, **Australia’s Medical Services Advisory Committee (MSAC)**—the official HTA body—issued a positive review for WES & WGS

- WES is reimbursed in **Japan** through the IRUD program
Societal Guidelines
ACMG recommends exome/genome sequencing (ES / GS) as a first- or second-tier test in patients with 1 or more congenital anomalies prior to 1 year of age or intellectual disabilities / developmental delay prior to age 18.

ES / GS leads to:

- Increased diagnostic yield in rare disease
- Awareness of broad spectrum of genetic variants
- Improved patient outcomes
- Expanded treatment and management
- Access to support networks for patients and families

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<thead>
<tr>
<th>Sequencing</th>
<th>WES/WGS</th>
<th>WGS</th>
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<tr>
<td><strong>Eligible Patients</strong></td>
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<td>Patients with one or more congenital anomalies prior to one year of age OR with intellectual disability with onset prior to age 18</td>
<td>Non-specific phenotype associated with intellectual disability and/or developmental delay; multiple congenital anomalies; clear clinical diagnosis associated with high level of genetic heterogeneity; previously negative WES or CMA</td>
<td>Any child &lt; 10 years with: facial dysmorphism AND ≥ 1 congenital structural anomaly; OR global developmental delay/ intellectual disability (moderate to severe); Test must be requested by clinical geneticist OR pediatrician following consultation with clinical geneticist</td>
<td>Patients w/ suspicion of significant monogenic disease associated with high degree of genetic heterogeneity; specific genetic tests have failed to provide a diagnosis; cases when WES/WGS is a more cost-effective approach than available individual gene/gene panels</td>
<td>It is recommended to introduce WGS analysis in a diagnostic setting when it is a relevant improvement on quality, efficiency and/or diagnostic yield</td>
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<tr>
<td><strong>Tier</strong></td>
<td>First or second tier test</td>
<td>First or second tier test</td>
<td><strong>Second tier</strong>: Negative routine blood tests if indicated, negative CMA required</td>
<td>First or second tier test</td>
<td>Not specified</td>
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<td><strong>Informed Consent and Pretest Counseling</strong></td>
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<tr>
<td>Set expectations, review benefits/limitations/ harms of testing such as limited disease-known associations</td>
<td>Discuss purpose of test, test limitations, possible results, possibility of secondary findings; possibility of data reanalysis</td>
<td>Explain possible outcomes to manage patient/parent expectations, address potential for incidental (secondary) findings, address possibility of certain types of insurance discrimination</td>
<td>Genetic counselling for the patient/ family should be undertaken and documented. A list of what should be discussed during the informed consent process is included in the document.</td>
<td>Pre-test genetic counseling should be performed prior to obtaining informed consent. This counseling should be performed by a qualified expert (ie. clinician, genetic counselor).</td>
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<td><strong>Reevaluation/ Reanalysis</strong></td>
<td>Value in reanalysis; frequency/strategy not specified</td>
<td>Not specified</td>
<td>In the event of a variant of uncertain significance, recommend reanalysis in 18 months, up to twice after the initial test is performed. Some situations warrant shorter reanalysis interval.</td>
<td>Requests for re-analysis should be initiated by a ref. physician based on an established policy; may involve re-testing rather than re-analysis, at discretion of laboratory. Further analysis of sequencing data through research may be an option.</td>
<td>Reanalysis should be triggered by the clinician and not by the diagnostic laboratory. Patients should be aware of and provide consent to reanalysis.</td>
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Continued Efforts
## What’s Next in Evidence-Generation

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<th>Yield</th>
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<tr>
<td>• Indication-specific diagnostic yields</td>
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<td>• Real-world evidence</td>
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<tr>
<td>• Multiomics (ie. RNAseq)</td>
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<td>• Long-read sequencing</td>
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<tr>
<th>Utility</th>
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<tr>
<td>• Change of management</td>
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<td>• Gene therapy/drug development</td>
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<td>• Economic utility</td>
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<td>• Personal utility</td>
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<th>Implementation</th>
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<tr>
<td>• Demonstrating what’s possible</td>
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<td>• Workflow impacts</td>
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<td>• Real-world implementation</td>
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<tr>
<td>• Education</td>
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iHope Global Network
Providing clinical whole-genome sequencing to patients in need

https://www.illumina.com/company/ihope.html
Looking Forward

Challenges for Whole-Genome Sequencing Implementation
Areas of focus for future test adoption

**Informatics**
- Supporting evidence for validated algorithms
- Data storage bottlenecks

**Interpretation**
- Incidental findings, variants of unknown significance
- Periodic data reanalysis

**Professional governance**
- Recommendations and guidelines for use
- Evidence for clinical utility and health economic benefits

**Access to testing**
- Access to testing facility offering clinical WGS
- Equity of care
- Payer reimbursement
Thank you
Pre-test Counseling for Genomic Sequencing

Test Characteristics

Result Types

Confidentiality and Privacy

Incidental Findings

Secondary Findings

Risks, Benefits and Limitations

Choice


Variant Filtering, Triaging and Classification

*Trio analysis*

- All Proband and Family Variant Calls
- Laboratory Quality Metrics
- Family Based Analysis
- Variant Classification Database
- Allele Frequency
- Variant Consequence
- Inheritance

List of candidate variants for clinical review and triaging
Systematic review of recommendations for reanalysis of next-generation sequencing data
Standardized guidelines for future studies relating to reanalysis

Key Takeaway
A meta-analysis of 29 articles determined that reanalysis of NGS data can improve diagnostic (dx) yield, yet there remains uncertainty regarding optimal timing for reanalysis. Recommendations are proposed for best practices in reanalysis and for minimum standards for future studies.

Method
- Systematic evidence review of publications from January 2007 to October 2021.
- Reanalysis defined as "bioinformatic examination of original sequencing data" in undiagnosed patients.
- Primary outcome was proportion of cases without a molecular dx after initial sequencing that reached a dx after reanalysis.

Findings
- **29 studies** (9419 undiagnosed patients) met criteria for review and included reanalysis data from GS (n=3), ES (n=23) and GS/ES (n=3) studies.
- Overall pooled dx yield from reanalysis was **10%**. Yields were higher when reanalysis occurred >24 months vs. those <24 months although this finding was not significant.
- Updates in literature/databases resulting in new gene discovery was most common reason for new diagnosis (62% of cases).
- Yield was significantly higher following reanalysis of ES data (11%) vs. GS data (4%) (p<0.01) and lowest in studies that limited reanalysis to known disease genes.