
# Copy number variants in a large cohort analysed with whole-exome sequencing: lessons for genetic diagnosis

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## Background

- Whole-exome sequencing (WES) enables the simultaneous analysis of all coding regions of the human genome.
- In routine molecular genetic diagnosis, genome-wide detection of copy number variants (CNVs) enabled the characterization of patients within cohorts with a broad phenotypic spectrum.

## Aim

To evaluate the efficacy of read depth-based CNV detection in routine diagnostics

## Methods

- 3,319 consecutive samples (2016 -2020)
- Capture kit:
  - Agilent's SureSelect Human All Exon (n=2819)
  - Twist’s Human Core Exome Kit (n=500)
- Sequencing: HiSeq4000 or NovaSeq
- Software: VarSeq (Golden Helix)
- Confirmation methodology: qPCR or MLPA

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## Results

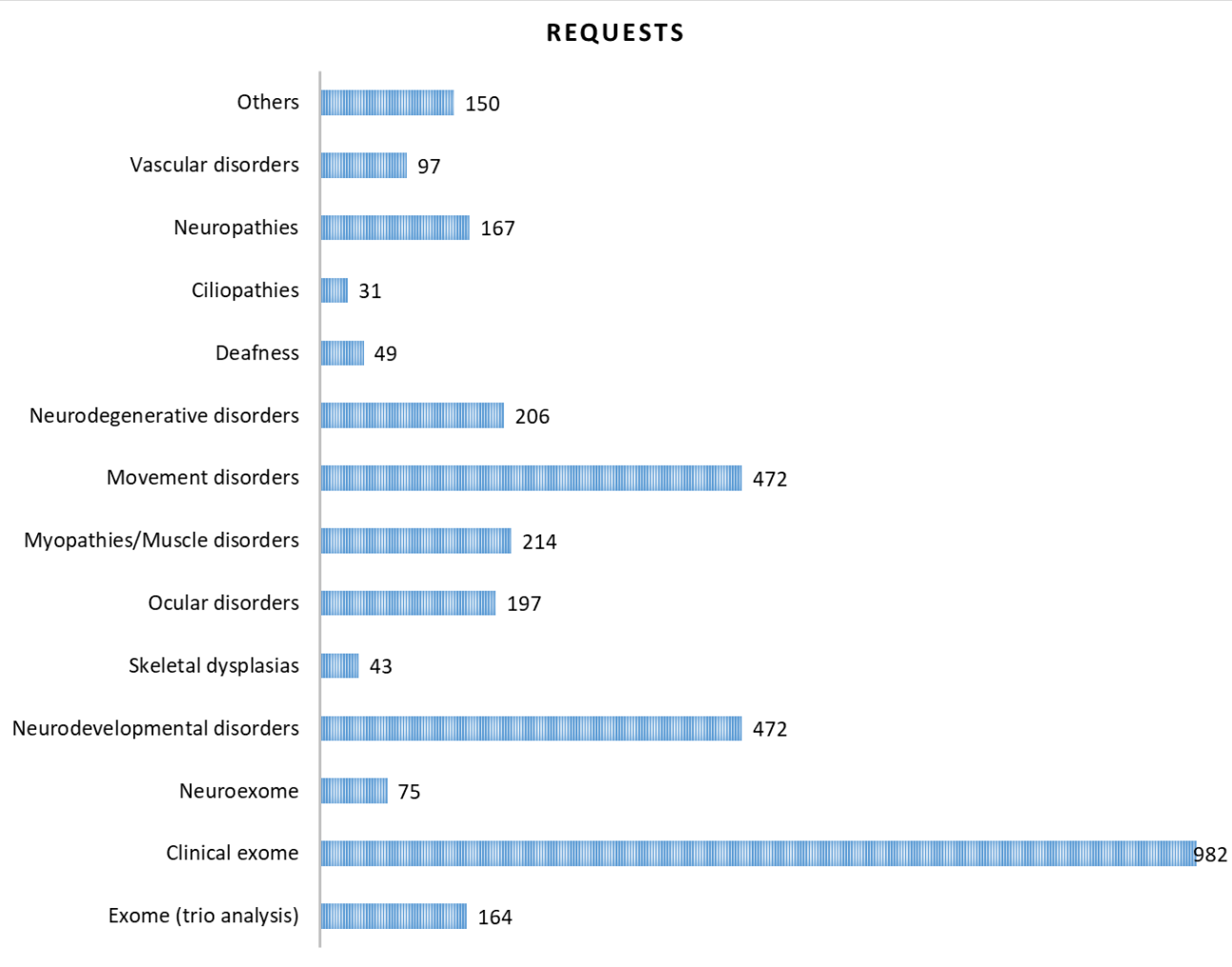


Figure 1 – Number of NGS-based genetic tests performed between 2016 and 2020 and subdivided by disease group.

The overall diagnostic yield attributable to CNVs was 4.6%

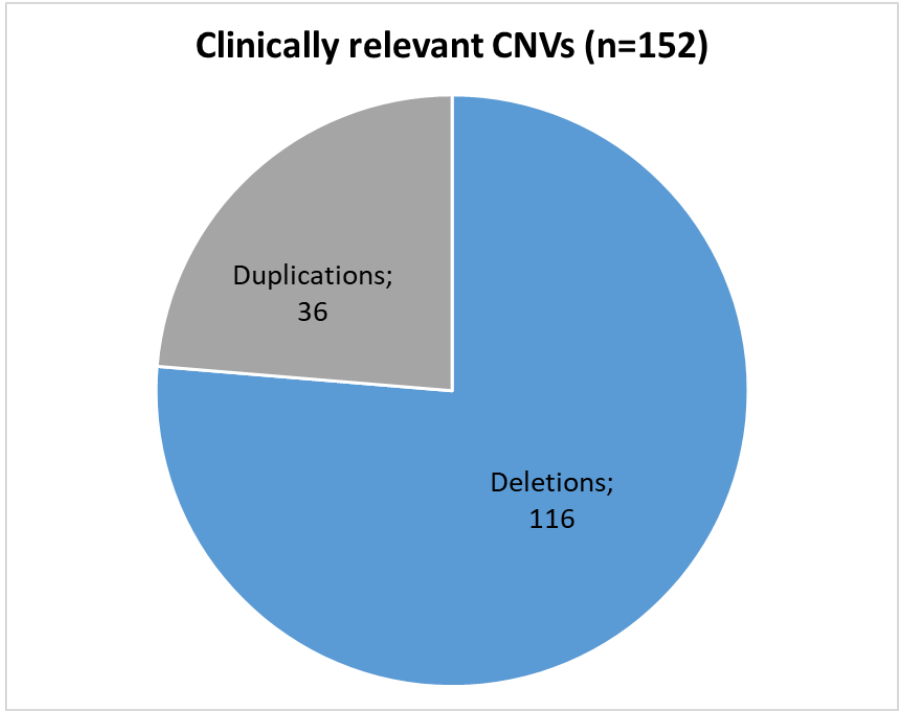


Figure 2 – Distribution of the 152 clinically relevant CNVs detected in this cohort.

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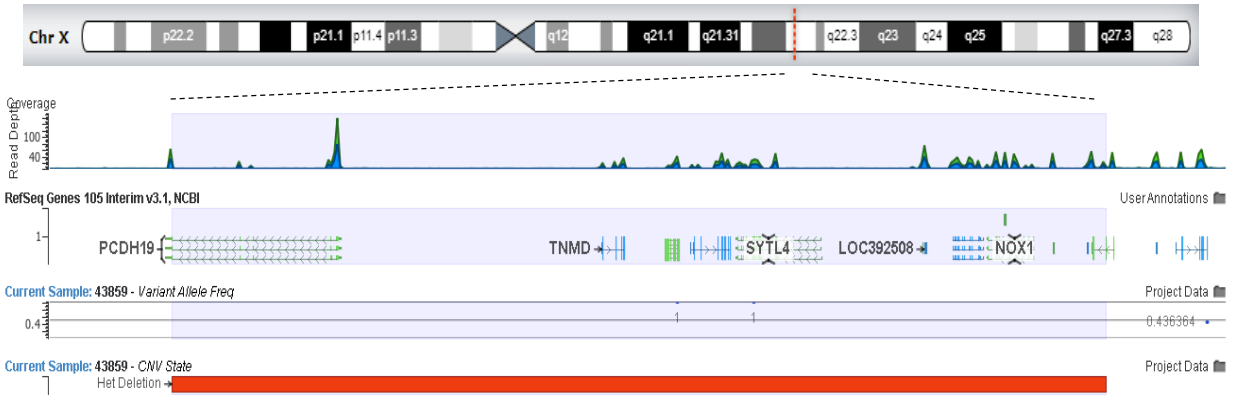
## Results

Table I – Distribution of the 152 detected CNVs by disease group and repective diagnostic yield.

| Disease group | Requests (total) | With CNVs | Diagnostic yield (attributable to a CNV) |
|---|---|---|---|
| Exome (trio analysis) | 164 | 7 | 4.3% |
| Clinical exome | 982 | 40 | 4.1% |
| Neuroexome | 75 | 3 | 4.0% |
| Neurodevelopmental disorders | 472 | 32 | 6.8% |
| Skeletal dysplasias | 43 | 3 | 7.0% |
| Ocular disorders | 197 | 15 | 7.6% |
| Myopathies/Muscle disorders | 214 | 7 | 3.3% |
| Movement disorders | 472 | 11 | 2.3% |
| Neurodegenerative disorders | 206 | 3 | 1.5% |
| Deafness | 49 | 4 | 8.2% |
| Ciliopathies | 31 | 3 | 9.7% |
| Neuropathies | 167 | 3 | 1.8% |
| Vascular disorders | 97 | 2 | 2.1% |
| Others | 150 | 14 | 9.3% |




## Patient:

- 1 year-old female patient
- Refractory seizures (onset at 4 months of age)
- Gene panel for epilepsies requested

## Genetic defect:

- The ~626.8 Kb deletion occurs at Xq22.1, affecting the *PCDH19* gene and other neighboring locus.
- Defects in *PCDH19* are associated with developmental and epileptic encephalopathy type 9 and affecting only females.

Figure 4 – Illustrative example of a CNV detected by depth-based analysis.

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## Results

Table II – CNV type and size distribution by panel/disease group.

| | CNV type | Total | | Intragenic | | Multiple genes | |
|---|---|---|---|---|---|---|---|
| | | Del | Dup | Del | Dup | Del | Dup |
| Disease group | Exome (trio analysis) | 2 | 5 | 1 | 4 | 1 | 1 |
| | Clinical exome | 32 | 8 | 27 | 6 | 5 | 2 |
| | Neuroexome | 2 | 1 | 2 | 1 | 0 | 0 |
| | Neurodevelopmental disorders | 17 | 9 | 15 | 5 | 2 | 4 |
| | Skeletal dysplasias | 3 | 0 | 3 | 0 | 0 | 0 |
| | Ocular disorders | 15 | 1 | 15 | 1 | 0 | 0 |
| | Myopathies/Muscle disorders | 6 | 1 | 6 | 1 | 0 | 0 |
| | Movement disorders | 9 | 2 | 8 | 2 | 1 | 0 |
| | Neurodegenerative disorders | 3 | 0 | 3 | 0 | 0 | 0 |
| | Deafness | 3 | 1 | 3 | 1 | 0 | 0 |
| | Ciliopathies | 3 | 0 | 3 | 0 | 0 | 0 |
| | Neuropathies | 2 | 1 | 2 | 1 | 0 | 0 |
| | Vascular disorders | 2 | 0 | 2 | 0 | 0 | 0 |
| | Others | 13 | 3 | 11 | 2 | 2 | 1 |

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## Discussion

***Yield:***

- Most of the CNVs detected in our cohort were intragenic
- Overall diagnostic yield attributable to CNVs was 4.6%
- By disease group:
  - 7.6% for ocular diseases
  - 6.8% for neurodevelopment disorders
  - 4.1% for clinical exome
  - 4.0% for neuroexome
  - 4.3% for WES trios
  - 3.3% for neuromuscular diseases
  - 2.3% for movement disorders
  - 2.1% for vascular diseases

***Pros:***

- WES allows the simultaneous detection of SNVs, InDels and CNVs
- Read depth-based CNV detection from WES data is a cost-effective add-on for diagnostic laboratories

***Cons:***

- High rate of false positives
- Requires confirmation by another methodology