Analysis of 37 / 65 muscle genes in 300 patients with neuromuscular diseases

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INTRODUCTION & METHODS

Neuromuscular diseases (NMDs) are clinically and genetically highly heterogeneous with more than 300 associated genes known to date making conventional molecular diagnosis challenging and expensive. Hence, we designed a targeted gene panel consisting of 37 (version 1) and 65 (version 2) genes associated with the most common types of muscular dystrophies and myopathies.

A total of 300 patients was analysed using the software GensearchNGS (PhenoSystems) after target enrichment of the selected muscle genes and next generation sequencing on a MiSeq desktop sequencer (Illumina). About 99% of all coding exons were covered >20x, the overall average coverage was >500x.

Table 1: Gene content of the myopathy gene panel, version 1:

ACTA1, ANO5, BAG3, BIN1, CAPN3, CAV3, COL6A1, COL6A2, COL6A3, CRYAB, DES, DMD, DNAJB6, DNM2, DYSF, EMD, FHL1, FLNC, FKRP, KLHL9, LDB3, LMNA, MTM1, MYOT, MYH7, RYR1, SEPN1, SGCA, SGCB, SGCD, SGCE, SGCG, SGCZ, SMCHD1, TCAP, TIA1, TTN

Table 2: Additional genes of the myopathy gene panel, version 2:

B3GALNT2, B3GNT1, CHKB, CPT2, DAG1, FKTN, GAA, GMPPB, ISCU, ISPD, LAMA2, LARGE, LPIN1, PNPLA2, POMGNT1, POMK, POMT1, POMT2, PYGM, SECISBP2, SIL1, TMEM5, TNNT1, TNPO3, TOR1AIP1, TPM2, TPM3, TRAPPC11, TRIM32

RESULTS

After removing recurrent sequencing artefacts and common variants (MAF>2%), we detected more than 4000 variants (~1400 different ones) in the 300 patients analysed, which we classified from benign to pathogenic (class 1-5). Among these were 35% missense, 29% intronic (9-20 bp flanking sequence), 24% synonymous, 7% potential/essential splice, 4% small deletions/insertions and less than 1% nonsense variants (Fig. 1). About 70% of the detected variants were classified as benign (class 1) or likely benign (class 2), approx. 20% as uncertain (class 3) and 3-4% as likely pathogenic or pathogenic each (class 4 and 5) (Fig. 2).

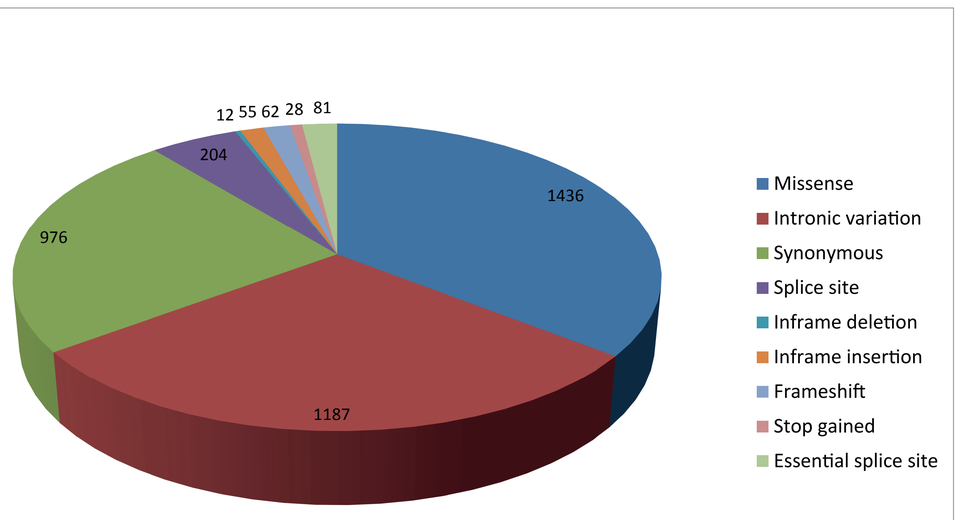

Figure 1:
Different variant types detected in 300 patients analysed by the myopathy gene panel.

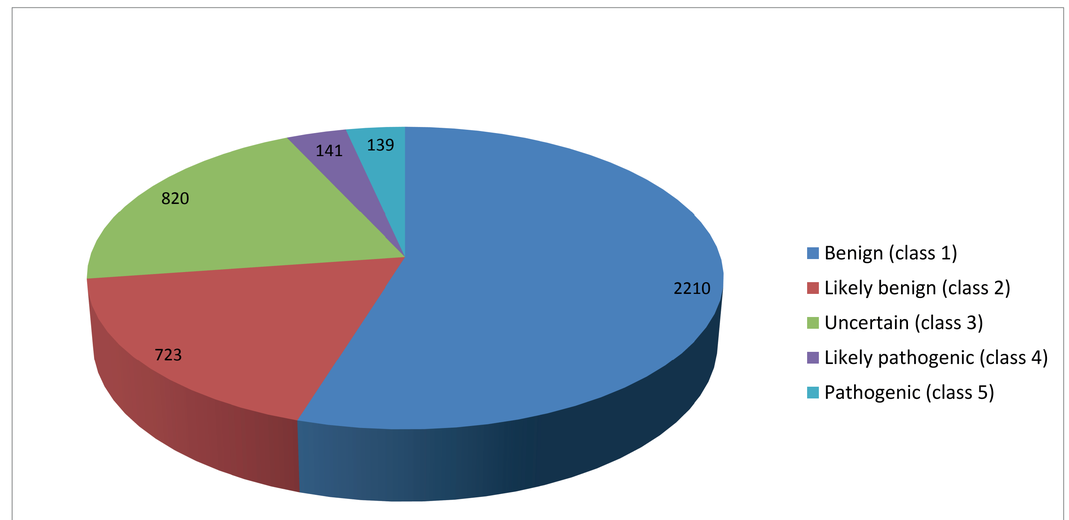

Figure 2:
Classification of variants detected in 300 patients analysed by the myopathy gene panel.

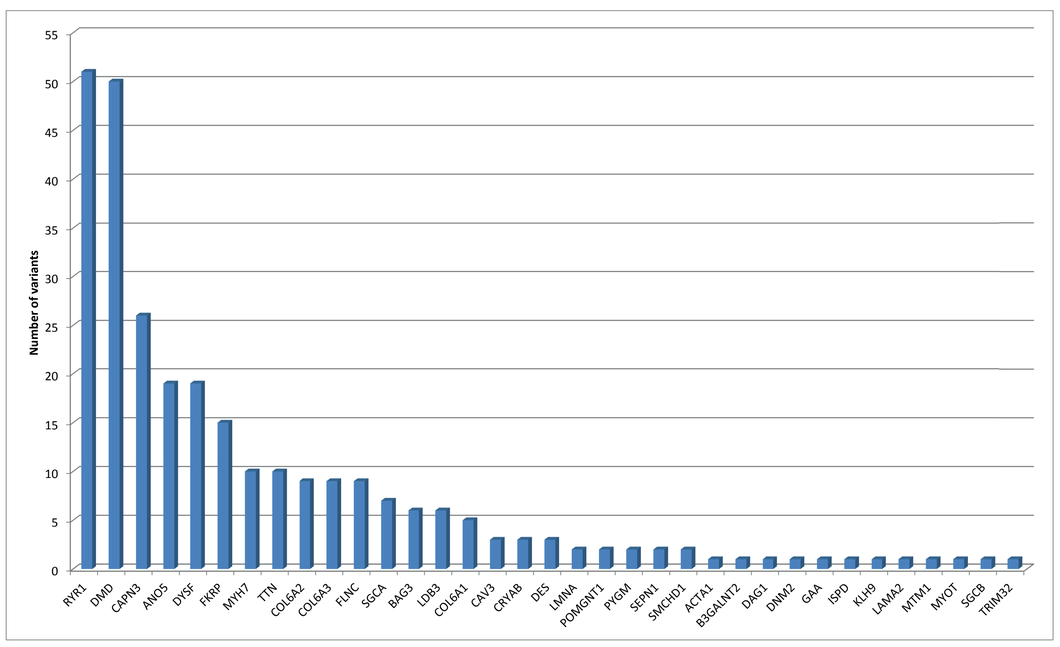

Figure 3:
Distribution of class 4 and 5 variants in 35 of the 65 muscle genes detected in 300 patients analysed by the myopathy gene panel. Only genes in which at least one class 4 or 5 variant was detected are displayed in the diagram.

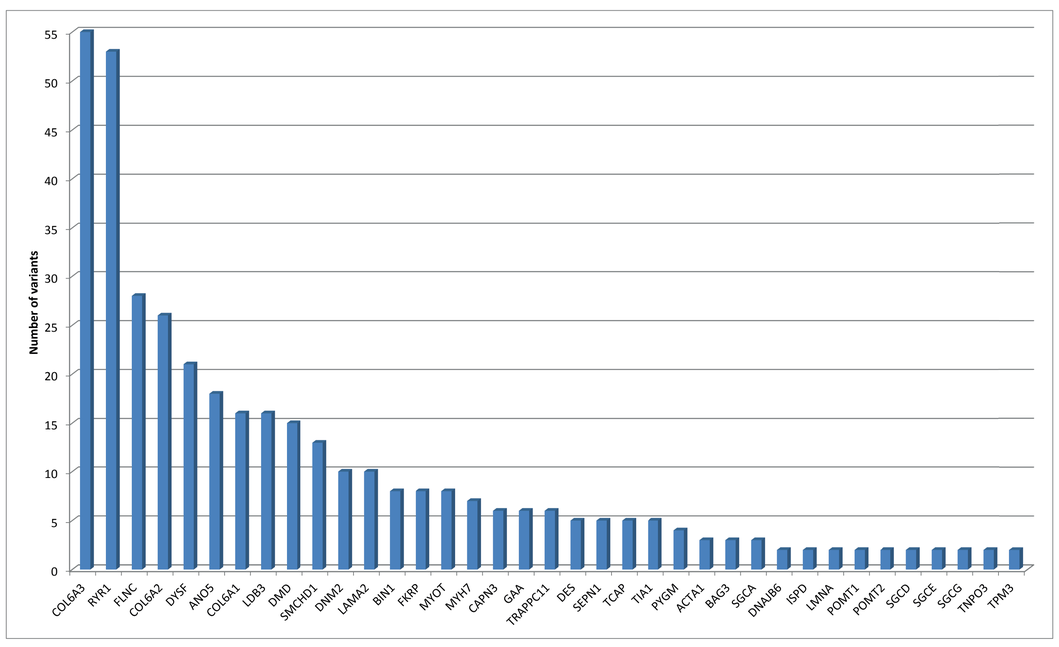

Figure 4:
Distribution of class 3 variants in 38 of the 65 muscle genes detected in 300 patients analysed by the myopathy gene panel. Only genes in which at least two class 3 variants were detected are displayed in the diagram. TTN with 416 class 3 variants is not shown.

CONCLUSION

At least one likely pathogenic or pathogenic variant (class 4 or 5; Fig. 3) was identified in 180 of the 300 patients suffering from different myopathies. This results in a detection rate of at least 60%. Most class 4 or 5 variants were detected in *RYR1* and *DMD*.

Furthermore, in almost all 300 patients analyzed by our muscle gene panel at least one variant of uncertain significance (class 3; Fig. 4) was detected which could be regarded as potentially associated with the myopathy.

Competing interests: None declared

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