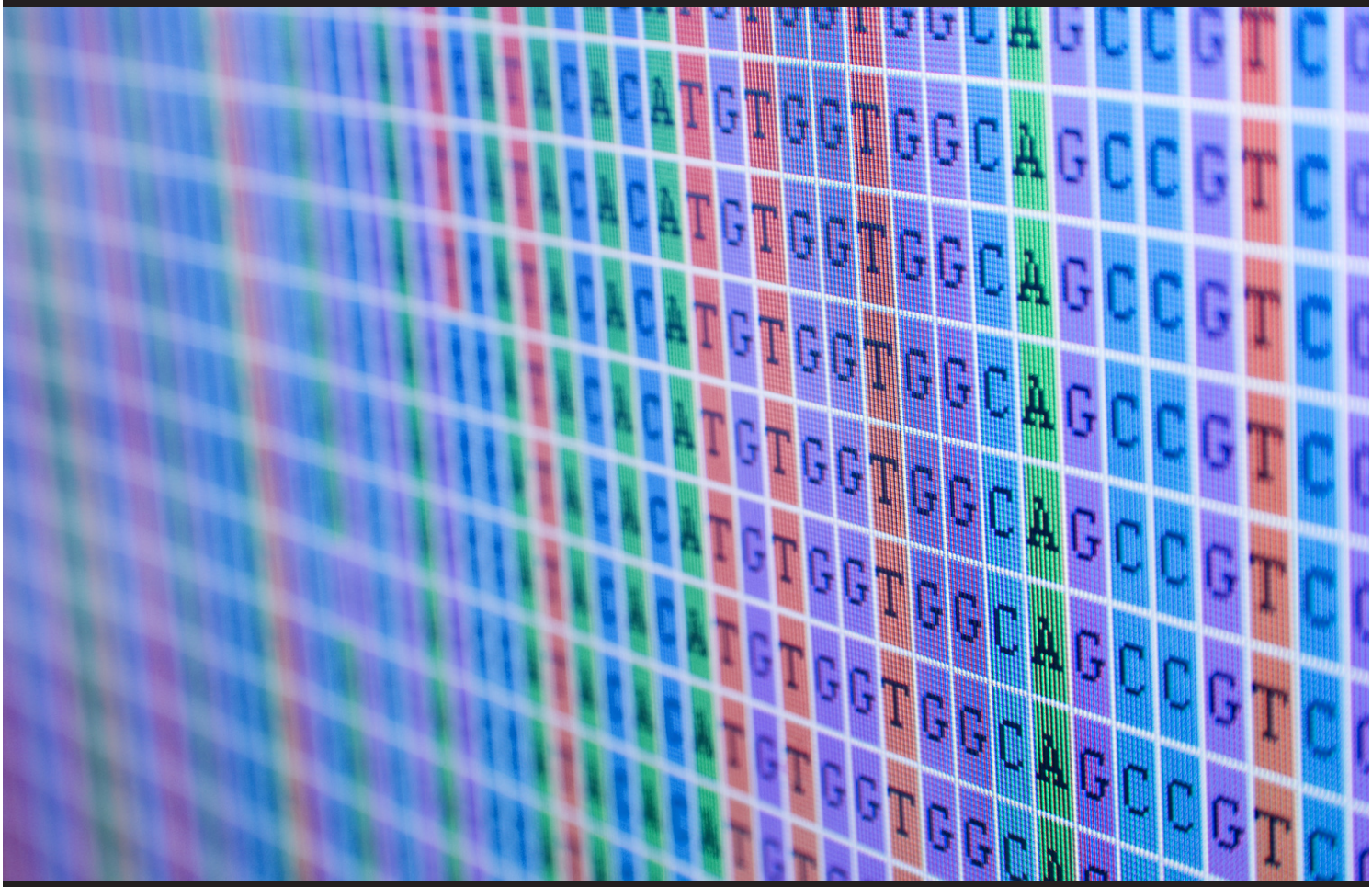
Slovenian Association of Medical Genetics
Slovenian Medical Association



7th Symposium of the Slovenian Association of Medical Genetics

NEXT GENERATION SEQUENCING IN CLINICAL PRACTICE

Book of abstracts

Ljubljana, 13th October 2017

Editors
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Publisher and year of publication
Slovenian Association of Medical Genetics, 2017

Image on the frontpage
Courtesy of GENETIC LITERACY PROJECT (https://geneticliteracyproject.org)

Kataložni zapis o publikaciji (CIP) pripravili v Narodni in univerzitetni knjižnici v Ljubljani
COBISS.SI-ID=293225216
ISBN 978-961-93050-5-8 (pdf)

Način dostopa (URL): http://www.szd.si/sekcije/medicinska-genetika/zdruzenje-za-medicinsko-genetiko

7th Symposium of the Slovenian Association of Medical Genetics

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Symposium presentations

T1.1

Next generation sequencing demands next generation phenotyping

Raoul Hennekam

T1.2

Sharing is caring – the significance of data sharing in the era of genomics

Ales Maver[1], Alenka Hodžić[1], Gaber Bergant[1], Tanja Višnjar[1], Karin Writzl[1], Borut Peterlin[1]

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Throughout history, exchange of information has been an essential momentum driving advancement of medical care. Sharing of findings is of particular importance in the field of human genetics, where we face the challenge of high multiplicity and complexity of findings on one hand, and their extreme rarity, on the other. As novel approaches, including whole exome and genome sequencing, are increasingly being used in routine genetic diagnostics, we identify a multitude of novel genetic variants of uncertain medical significance on a daily basis. Because functional assays are often too time-consuming and expensive to perform routinely, identification of other individuals carrying the same genetic alteration commonly remains the single path to clarification of genetic variants' medical impact.

In the past, genetic variants were predominantly shared through scientific journals and several disease-specific databases. However, this mode of sharing has been too slow and too restrictive to compensate the exchange of data generated using current sequencing approaches. Publicly available repositories of variants (ClinVar, LOVD) have now been established as platforms where institutions can directly deposit their variants along with their medical impact assertions. This mode of sharing offers several benefits, including faster time to release variant information, facilitation of sharing findings of uncertain significance and comparison of assertion consistency among various institutions.

When diagnosis of a patient and their medical management is dependent on resolving the effect of a novel variant, there is a need to actively search for patients with matching genotype and phenotype information. To enable such exchange, an orchestrated Matchmaker Exchange effort within the Global Alliance for Genomics and Health (GA4GH) has been established to collect and connect multiple repositories with genetic and phenotypic information of patients. These repositories operate as a federated network, which enables identification of similar cases in institutions across the globe. This platform has now been used successfully in both, clarification of variants' significance and identification of novel genes for Mendelian disorders.

Historically, data sharing has been regarded to be mostly of scientific interest and the variants were deposited in rather inert databases and repositories. Nowadays, understanding of genetic variants has become so dependent on the process of data sharing, that it should be regarded as an integral step in provision of diagnostics for patients with mendelian disorders.

T1.3

Applying a quantitative approach to variant interpretation in inherited cardiac conditions

Roddy Walsh
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Background: Cardiomyopathies are a group of Mendelian genetic diseases that affect approximately 1 in 200 people and are the leading causes of both sudden cardiac death in the young and heart failure requiring transplantation. Genetic testing is recommended for both hypertrophic cardiomyopathy (HCM) and dilated cardiomyopathy (DCM) and is widely adopted, with the primary purpose of enabling cascade screening in family members to identify individuals at risk of disease, as well as those free from risk and therefore released from ongoing clinical surveillance. However several issues limit the effectiveness of genetic testing in these conditions – dubious gene and variant associations in the literature, strict variant interpretation guidelines leading to an under-calling of pathogenic variants and a bias in favour of individuals with well-characterised variants and populations that have been extensively studied.
Methods: By utilising large case and reference population cohorts, we can apply more quantitative approaches to gene and variant pathogenicity in cardiomyopathies. Applying disease-specific thresholds for variant population frequency, we have compared the burden of rare variation in large clinical cohorts in HCM and DCM with the background population rate in ExAC. We define the etiological fraction (EF) as the likelihood that a rare variant in a patient in disease-causing and use this to estimate the interpretability of genes, variant classes and regions within genes. We propose adaptations to ACMG guidelines that apply this quantitative approach and enable more thorough and unbiased variant interpretation.
Results: We observed a significant case excess of rare variation in HCM and DCM genes that have well characterised through linkage studies but little or no excess in more recently implicated genes, casting doubt on their role in disease. By re-evaluating the published evidence for genes implicated in HCM, we conclude that the majority are not associated with the disease and novel, non-sarcomeric genes are likely to be causative in less than 2% of cases. We identified regions within 5 key HCM genes that have an EF≥0.95 and, when applying our adapted ACMG guidelines, lead to a 14-20% increase in the yield of actionable variants in HCM.
Conclusions: Robust quantitative and statistical analysis of large case and control genetic datasets can clarify which genes are likely to yield actionable results when included in clinical genetic testing and identify variant classes with high prior probabilities of pathogenicity, increasing the yield of high confidence pathogenic variants.

T1.4

High-Throughput Population-Wide Genetic Screening for Inborn Genetic Disorders: Are we there yet?

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A great progress in the field of DNA/RNA sequencing technology of the last decade has boosted the genomic science and significantly influenced the field of medical genetics. With the development of high-throughput DNA sequencers, next generation sequencing (NGS) technology and novel analytical algorithms as well as the release of huge public databases of genetic variant's allele frequency in general population (ExAc, GnomAD, …) the paradigm of medical genetics dramatically changed and shifted from diagnosis driven to phenotype driven analytical approach. At the same time, the price to generate genomic data significantly dropped and consequently the possibility of potential universal preventive (neonatal) genetic screening has become a reality.

Nevertheless, the specifics of universal disease screening (UDS) in neonatal or paediatric population have to be addressed before actually introducing one into the healthcare system. The issues regarding UDS range from technical to ethical, including the question of specific healthcare system capacity to cope with newly diagnosed patients as well as the question of funding such an extensive high-tech operation.

The first newborn screening program was established in the USA (phenylketonuria in 1962) and gradually expanded to cover 31 primary and 26 secondary conditions. The majority of testing is done by mass spectrometry technology, enabling fast and relatively cost-effective screening program. There is quite a significant variability in the established newborn screening programs in European Union where the regulation is left in hands of each member state alone. For example, in Austria the universal newborn screening covers 25 metabolic disorders, 2 hormone disorders and cystic fibrosis. At the same time, newborns in Slovenia are screened only for one metabolic and one hormone disorder and 5-years old children for familial hypercholesterolemia.

Consequently, a great effort of medical experts to expand the newborn screening in Slovenia resulted in the finalization of the expanded newborn screening program with 13 new disorders that will come in to life in 2018. Additionally, a study to evaluate the potential of NGS and genetic testing as a 2nd tier test of expanded newborn screening program has been performed. The results supports the introduction of NGS into the newborn screening program as a 2nd tier test to clarify potentially vague results of biochemical tests.

The results, encountered issues and conclusions of abovementioned study will be briefly presented, together with legislation, ethical and technological issues regarding introduction of potential genetic standalone screening program.

T2.1

Defective actin arp2/3 activator function caused by a arpc1b mutation results in a novel Wiskott- Aldrich syndrom like immunodeficiency

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Background:
Polymerisation of actin filaments is initiated by actin nucleators. Among them Arp2/3 complex has ability to form branched actin networks and is regulated by members of the Wiscott-Aldrich syndrome protein(WASp) family. Mutation in WASp gene can cause immunodeficiency characterised by recurrent bacterial infections, eczema, thrombocytopeniaand autoimmune diseases.
We present three related patients with WAS features and a mutation in a *ARPC1B* gene(Arp2/3 activator subunit).

Methods:
All patients (2 females,1 male–relatives) presented in neonatal period with eczema, thrombocytopenia and bloody diarrhoea. Later they developedrecurrent bacterial infection of the lung and skin, therapy resistant inflammatory bowel disease, various autoimmune features(AI vasculitis,AI thrombocytopenia,perniciousanaemia) and allergic reactions. They have mild cellular deficiency, slightly decreased phagocytic assays, decreased IgM and elevated IgA and IgE antibodies.
Whole exome sequencing was performed on an index patient.
A simple, rapid and very specific functional test to evaluate Arp2/3 complex function was developed, consisting of *in vitro* fMLP stimulation of mononuclear cells and flow cytometric detection of intracellular polymerised actin with fluorescinated phalloidin.

Results:
We identified a missense mutation in novel gene *ARPC1B*(Arp2/3 activator subunit) that was predicted disease causing with several in silico prediction tools.
Median fluorescence intensities(MFI) of FITC-phalloidin stained actin in monocytes and neutrophils as evaluated by flow cytometry are shown in table 1.

Conclusions:
We report three patients with novel WAS-like immunodeficiency caused by mutation in ARP2/3 activator subunit.
Functional fMLP/phalloidin test can efficiently discriminate homozygous symptomatic patients from asymptomatic heterozygous carriers and can be used as a screening test for actin-polymerisation defects.

T2.2

High germline *BRCA* mutation detection rate among epithelial ovarian, fallopian tube and primary peritoneal cancer patients in Slovenia

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BACKGROUND: In Slovenia, olaparib has been registered for treatment of relapsed platinum-sensitive *BRCA*-mutated high-grade serous epithelial ovarian, fallopian tube and primary peritoneal cancer since February 2016. Therefore, all patients with newly diagnosed and relapsed disease are referred to our Cancer Genetic Clinic and offered genetic counselling and testing, regardless of their family history. According to the literature, *BRCA* mutation detection rate of around 22% was to be expected in these patients.

AIM: The aim of this study was to analyse the participation rate in genetic counselling and testing and to assess germline *BRCA1/2* mutation detection rate among referred patients.

METHODS: We included all patients with epithelial ovarian, fallopian tube and primary peritoneal cancer who were referred to our Cancer Genetics Clinic for genetic counseling and testing from October 2014 till September 2016. Genetic testing was performed with the next-generation sequencing using Illumina's TruSight Cancer sequencing panel.

RESULTS: In the two-year period 299 patients were referred to our Clinic. 258/299 (86.3%) patients contacted us and opted for *BRCA* testing, 39/299 (13%) never made contact and two (0.7%) contacted us for counseling but declined genetic testing. *BRCA1/2* mutation detection rate was 36% (93/258 pts). Among *BRCA* positive patients, 71/93(76.3%) carried a *BRCA1* and 22/93 (23.7%) a *BRCA2* mutation, respectively. In 6% (16/258) we identified mutations in other genes from the panel. Out of 258 tested, 165 (64%) patients had positive family history, and among those in 44.8% (74/165) the *BRCA1/2* mutation was detected. In the remaining 36% (93/258) of patients with negative family history, 18.3% (17/93) had the *BRCA1/2* mutation. Among 134 patients, who were tested at the time of ovarian cancer diagnosis, 33.6% had the *BRCA1/2* mutation detected.

CONCLUSIONS: Genetic testing for germline *BRCA1/2* mutations in patients with ovarian, fallopian tube and primary peritoneal cancer yielded a very high mutation detection rate, regardless of family history and disease setting. Further research is needed to explore these interesting findings.

T2.3

The majority of persons with ABCA4 disease-associated genotypes do not present with visual disability

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Purpose: The population variant data suggests higher frequency of disease-associated variants in *ABCA4* than expected considering the rarity of disease. To surmount the lack of data on the true prevalence of visual loss due to *ABCA4* mutation, we assumed full penetrance of those with nullizygous genotypes and determined which other genotypes (if any) were under-represented in patients.
Methods: Hypothetical distributions of genotypes generated from GnomAD and NIHR-Bioresource allele frequency data were compared with those observed in patient cohorts (397 probands from UK; 166 from Netherlands). Phenotypes were analyzed using fundus autofluorescence imaging (FAF).
Results: There was significantly skewed distribution of nullizygous (N/N), hemizygous (N/M) and bialleic missense (M/M) genotypes in patients with at least 11-fold paucity of missense genotypes. Two specific genotype subclasses occurred in homozygous but never in the compound heterozygous state. These patients had different patterns of retinal pathology on FAF.
Conclusions: Results suggest low penetrance of *ABCA4* genotypes. *Cis* and *trans* modifying factors are likely and a reevaluation of the haplotypes is warranted. Hardy Weinberg Disequilibrium of specific allele subclasses suggests there is reciprocal compensation of different dysfunctions of ABCA4 protein in some cases.

T2.4

Mosaicism for a *NF1* mutation as a secondary finding on multi-gene panel testing

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Genetic testing for hereditary cancer predisposition syndromes is often performed using a next-generation sequencing (NGS) based approach. With its higher diagnostic yield and shorter turnaround time it is quickly replacing other sequencing methods. NGS enables the simultaneous analysis of many disease-causing genes but can lead to the discovery of secondary and unsolicited findings, even when using a targeted gene panel.

We report a case of a 29-year-old patient with adenocarcinoma of the rectum, referred to our clinic for genetic counselling. He had had a gastric operation in early childhood and a history of gastritis but no other health issues. His family history was unremarkable with the exception of two paternal uncles who developed colorectal carcinoma, aged 42 and 57. Due to early-onset disease and positive family history he was offered testing with a core panel of genes associated with a predisposition to colorectal carcinoma. On pre-test counselling the patient was informed of the potential for secondary findings. Testing was performed using Illumina's TruSight Cancer sequencing panel and showed no pathogenic variants in the core gene panel, but revealed a known pathogenic variant c.6858+1G>A in *NF1*. As the variant was present in 33% of reads and the patient did not fulfil clinical diagnostic criteria for neurofibromatosis type 1 (NF1), mosaicism appeared the most likely explanation. Further analysis of various tissue samples showed the presence of the *NF1* variant in approximately 30% of alleles in tissues of mesodermal origin and 10% of alleles in most tissues of endodermal origin. It was not detected in DNA extracted from buccal swabs which implies it might be absent from tissues of ectodermal origin, thereby explaining the absence of NF1 features in our patient. The variant was not detected in the patient's adenocarcinoma.

Our case highlights the issues that arise on detection of secondary findings using NGS. Mosaic NF1 is usually diagnosed based on phenotypic findings and the risks and possible surveillance programs for these patients are poorly defined. Although the possibility of NF1-associated complications and gonadal mosaicism seems low in our case neither can be ruled out, necessitating further evaluation. Additional recommendations on reporting secondary findings could help unify laboratory reporting policies and possibly improve patient management.

T2.5

Next Generation Sequencing in the molecular diagnosis of multiple cardiovascular disorders

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Introduction: Cardiovascular disorders are one of the main cause of death worldwide and are a heterogeneous group of diseases that involve heart and blood vessels. There are many different cardiac conditions that have an underlying genetic contribution. Some are straight forward Mendelian disorders whereas others have a more complex genetic involvement. Inherited cardiac conditions can be discovered and diagnosed in a patient based on multiple different factors. In all cases of a suspected genetic disorder, genetic testing is an important tool to help clarify and specify the precise diagnosis, and may be especially useful in cases of borderline clinical findings. Recently, next generation sequencing (NGS) has been successfully applied into the clinic to find causal mutations and thus confirm the clinical diagnosis in patients suspected of hereditary cardiac disorders.

Material and Methods: NGS analysis of genomic DNA was performed using the Illumina TruSight Cardio panel, targeting 174 genes involved in several cardiac disorders, on MiSeq platform. We performed two runs including 24 patients with different cardiac phenotypes. RFLP method or ARMS PCR method or Sanger sequencing were used to support pathogenicity of potentially causal mutations. Variant Studio (Illumina) software and free-access tools and databases were used for analysis and interpretation of the NGS data.

Results: NGS achieved 300x average of mean region coverage depth and yielded a coverage >10x in 99.3% targeted regions. In 3 patients pathogenic/probably pathogenic variants were found. The variants of unknown significance (VOUS) were identified in 5 patients. On the other hand, the genetic cause of the pathology was not found in 16 patients.

Conclusions: NGS allows fast and efficient variant detection to improve clinical management and family counseling. It means that a single test may identify the causative gene mutation in someone with a heart condition thereby allowing their relatives to be easily tested for the same gene mutation. However, the amount of data obtained during the NGS analysis highlights the need of precise clinical definition and better strategies for the determination of the pathogenicity of the identified variant.

T2.5

Telomere length and arterial pulse wave velocity in children with hypercholesterolemia

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Telomeres are repetitive, non-coding DNA sequences (TTAGGG) at the end of chromosomes and their length is affected by several factors. In patients with hypercholesterolemia (HH), one of them is the prolonged exposure to oxidative stress associated with inflammation. High lipoprotein(a) (Lp(a)) concentration in HH patients is associated with impaired endothelial function and coronary artery calcification. A number of factors for risk assessment have been studied, including the arterial pulse wave velocity.

Our hypothesis were that children with HH and high Lp(a) have shorter telomere length (TL) and greater pulse wave velocity compared to children with HH and lower Lp(a).
The study included 58 children (aged 5-8 years at the begging of the study, 5 with mutations in *APOB* gene, 23 with mutations in *LDLR* gene, 21 with wild type mutations and 9 without results of the genetic test) referred to the University Children's Hospital Ljubljana through the national screening program for HH. In 32 children Lp(a) values were < 100 mg/L in 26 they were > 500 mg/L. Relative TL was determined with modified Cawthon's method of monochrome multiplex quantitative real-time PCR (MMQPCR). To estimate arterial stiffness pulse wave velocity of the aorta was determined with Complior Analyse®.

Groups did not differ regarding age, gender ratio and total cholesterol, HDL, LDL, triglycerides and present mutations. Relative TL was shorter in the group with a higher Lp(a) ($p = 0.0011$; unpaired t-test with Welch correction) were also hsCRP levels where higher ($p=0,0195$; unpaired t-test with Welch correction). Pulse wave velocity tended to be lower in the group with Lp(a) below 100 mg/L, it however didn't reach statistical significance.

Children with HH and higher Lp(a) values have shorter relative TL and higher hsCRP levels comparing to those with lower Lp(a) levels. This is indicating that as early as in childhood, higher levels of the oxidative stress associated with inflammation due to high cholesterol levels are resulting in shorter telomeres however, arterial stiffness at that stage was implied but not yet significant.

T3.1

Nanopore sequencing of human mtDNA

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Easy, fast and accessible DNA/RNA sequencing is a fundamental basis for a genetic research. The nanopore sequencing technology is a novel approach to sequence DNA/RNA through a protein or solid-state nanopore. It has the potential to achieve four gold standards of the DNA sequencing (high accuracy, long read length, high throughput and low cost) and make sequencing even more accessible. The aim of this study was to test and evaluate the performance of the nanopore technology by sequencing a known whole mtDNA sequence using the MinION (Oxford Nanopore Technology Ltd., UK). The mtDNA was amplified from the whole-blood DNA sample by the long-range PCR in two segments (9067bp and 11172bp). The nanopore sequencing library was prepared with a modified protocol, roughly following manufacturer's instructions, followed by two hours long sequencing run on the MinION. The obtained data was analyzed in the Galaxy environment (*http://galaxyproject.org)*. In this short sequencing run, 2116 successful paired-end reads were generated. 99.01% were mapped to the reference sequence (hg_g1k_v37) and 1792 of them mapped to the reference mtDNA sequence, resulting in an enrichment of 85.5%. The average coverage of mitochondrial genome achieved by sequencing was 492x with a mean read length of 8323 nucleotides and mean base-quality Phred score of 22. Nevertheless, the sequencing error rate was 12.9%. The accuracy of variant calling was significantly influenced by the variant caller used. The best results were achieved using the consensus sequence generated by IGV tools, which applies the variant calling method described by Cavener, with 87.5% sensitivity and 13% false discovery rate. Interestingly, when using SAMtool variant calling pipeline and different minimal quality value cutoffs (Q30, Q27, Q25, Q20), the results were far worse with the sensitivity as low as 17.6%, 25%, 31.3% and 43.8%, respectively, and false discovery rate of 40%, 80%, 87.5% and 92%, respectively. In conclusion, we successfully sequenced the mtDNA by using the nanopore sequencer, but the reliability to identify genetic variants is still lacking, especially when the potential heteroplasmy of mitochondria is taken into the account. With the improvement of base calling, alignment and variant calling algorithms optimized for long reads, the potential of this technology in a diagnostic setup will be greatly enhanced.

3.2

Analysis of mutational burden in exome sequencing data of patients with multiple sclerosis

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BACKGROUND

Multiple sclerosis (MS) is a debilitating neurological disease affecting young adults. It is considered to be an autoimmune multifactorial disease with its heritability estimated at 15%. While it's etiology remains unexplained, several recent studies have implicated the role of inflammasome and other elements of the innate immune system. Our aim was to investigate the mutational burden of rare variants in the inflammasome pathway in patients with MS.

METHODS

We analyzed whole exomes of 43 sporadic, 49 familial MS patients, and of 92 age and sex matched controls. Raw data was analyzed according to GATK best practices (Joint genotyping with subsequent Variant Quality Score Recalibration). Rare missense variants were collapsed on NLRP1/CASPASE1 centered panel of 50 genes and annotated with CADD pathogenicity prediction software.

RESULTS

Familial MS cohort is enriched for the variants with the highest pathogenicity predictions (CADD > 20) compared to controls (chisq, p=0.0004). Moreover, the enrichment steadily increases when the predicted pathogenicity inclusion threshold for the variants is raised. This trend is not observed with randomly generated gene panels of the same size.

CONCLUSIONS

Our analysis points to a significant contribution of the burden of rare functional genetic variants in NLRP1/CASPASE1 pathway to the etiology of MS in patients from families with multiple affected members.

T3.3

Global differential expression of small RNAs in the muscle tissue of ALS patients.

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Amyotrophic lateral sclerosis (ALS) is a late onset disorder leading to progressive and lethal skeletal muscle atrophy, resulting from death and dysfunction of motor neurons. Small RNAs, including microRNAs (miRNAs), can serve as important regulators of gene expression and are importantly involved in regulation of many cellular processes. Several muscle miRNAs or myomiRs are dysregulated in various other neuro-muscular disorders, and/or have shown promise for therapeutic use in cellular and animal models of ALS; however the global expression of miRNAs in muscle tissue of ALS patients has not yet been determined. Following small RNA-Seq from muscle tissue of ALS patients and controls, we performed differential expression analyses of small RNAs. The identified snoRNAs, mtRNAs and other small RNAs suggest novel molecular links between insulin signalling and ALS. The identified miRNAs further support the hypothesis that muscle tissue is undergoing active reinnervation/compensatory attempts in ALS thus providing targets for further research and therapy development.

T3.4

Advanced genomic analyses of Slovenian children with Early Onset Schizophrenia

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Introduction: Schizophrenia is complex psychiatric disorder with polygenic inheritance. Disorder is rare in children; prevalence is estimated to be 1 in 1,000. Early Onset Schizophrenia (EOS) is classified as a disorder where symptoms are present after the age of 13 years. Before that age, children are diagnosed with Very Early Onset Schizophrenia (VEOS). Preliminary data suggest that EOS and VEOS have higher genetic liability to the disease. Therefore, we preformed genetic testing using Next Generation Sequencing (NGS) for schizophrenia-associated single nucleotide variants (SNVs) in a small group of Slovenian patients diagnosed with EOS.
Material and Methods: A NGS analysis was performed with Illumina TruSight One (TSO) kit on Miseq platform for 30 patients. Using the on-line available tools and literature, a panel of 168 schizophrenia-associated genes was created which were screened for disease causing mutations in our set of patients. Data interpretation was performed with Illumina Variant Studio and on-line available tools and databases.
Results: An average of 40x mean coverage depth was achieved for each patient and accuracy of 94% was shown to be achievable at 10x average coverage. In 70% (21/30) patients no known pathogenic variants were identified for the selected genes. In the remaining 30% (9/30) variants of unknown significance (VOUS) were identified.
Conclusions: The performed NGS analysis did not shown any of known pathogenic variants, which was not surprising, considering of the polygenicity of the disorder and a small sample size. In nine patients the identified variants could be involved in aetiology of EOS. However, according to the American College of Medical Genetics (ACMG) guidelines the variants are classified as VOUS, due to their relative high population frequency or the unknown impact of the variant on the biological role of the gene. Increasing the number of patients and building the reference database of the Slovenian populations should contribute to a better understanding of the meanings of the identified variants in patients.

T3.5

Towards using Literature-based Discovery for NGS Results Interpretation

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Next generation sequencing (NGS) has the potential to revolutionize clinical genetics. However, there are still numerous challenges when interpreting NGS results. Here we present our preliminary research in using literature-based discovery (LBD) paradigm to improve the interpretation of NGS results. The goal of LBD is to generate novel hypotheses by analyzing the literature and optionally other knowledge sources. LBD uses either of two basic approaches: open discovery and closed discovery; both are based on a paradigm of three related concepts: X, Y, and Z. In open discovery only the starting concept is known. In closed discovery both the starting concept (X) and the end concept (Z) are known, and we want to find intermediate, linking concepts (Y) that may help explain the relationship between X and Z.
For this work we selected closed discovery LBD as more suitable. Here is a short description of the algorithm. We deal with a single patient data at a time. The input are two sets of data for each patient; set X contains the observed human phenotype ontology (HPO) terms, and set Z contains the genes with mutations as found by NGS. The output of the algorithm is a set of intermediate concepts Y that link the genotype Z to the phenotype X. These Y concepts should provide a hypothesis that explains the mechanisms that link the genotype to the phenotype. Our algorithm is meant as a discovery support step in a more general NGS data processing pipeline. We generate hypotheses (explanations), but a knowledgeable human expert is needed for the critical evaluation of these hypotheses.

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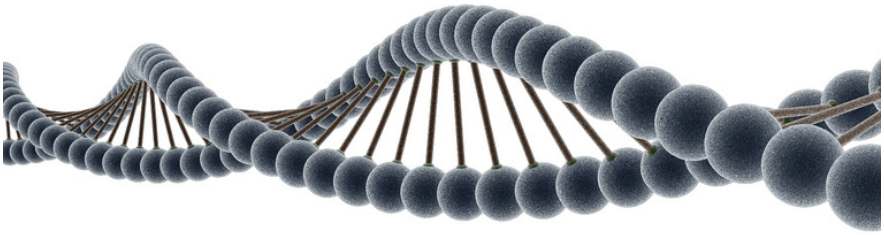

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