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Oral Presentation Awards

1st Place

Confirmation of *PATL1* Gene as Neurodevelopmental Disease Gene by Fruit Fly Model

Berfin Dağ, Mert Tufan, Abdullah Sezer, Arzu Çelik Fuss

2nd Place

Evaluation of the Effectiveness of Combined Treatment with the PD-L1 Inhibitor Atezolizumab and the C-MET Inhibitor Crizotinib in MCF-7 Breast Cancer Cells

Neslihan Haklıgür, Ahsen Güler, Mevlüde İnanç

2nd Place

Importance of SNP-Based Microarray Technology in Disorders of Sexual Development Workup; Lessons Learned from a Male Case with 46,XX Karyotype

Ali Torabi, Talha Laçin, Burak Aktaş, Ali Çiçekli, Özkan Bağcı, Ebru Marzioğlu Özdemir, Nadir Koçak, Tülin Cora, Muammer Büyükinan, Muhammed Emin Sarı, Gülsüm Türkoğlu

3rd Place

Deletion of Multiple Exons of KIF1C Detected by Next Generation Sequencing Associated with Spastic Ataxia 2

Ebru Tunçez, Oğuzhan Bahadır

3rd Place

Relationship Between *GSTP1, XRCC1, ERCC1, MTHFR TSER* and *DPYD* Gene Polymorphisms and Progression-Free Survival in Colorectal Cancer Patients Received FOLFOX Treatment

Ege Rıza Karagür, Atike Gökçen Demiray, Hakan Akça

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Poster Presentation Awards

1st Place

Legius Syndrome with a Preliminary Diagnosis of NF1-like Syndrome: Case Report

Büşra Saruhan, Momen Kanjee, Ziya Bulduk, Dilara Aydemir, Çiğdem Yüce Kahraman, Abdulgani Tatar

2nd Place

Hereditary Hyperekplexia: Three Patients from Kayseri, Middle Anatolia and Three Different Genetic Findings by Different Methodology

Maide Korkmaz, Aslıhan Kiraz, Hakan Gümüş, Hüseyin Per, Munis Dündar

2nd Place

Duplication of 1q21.3q25.3 in a Newborn with Multiple Congenital Anomalies

Leyla Nur Yılmaz, Tamer Güneş, Hilal Akalın, Aslıhan Kiraz, Yusuf Özkul, Munis Dündar

3rd Place

Case Report: Patient with Merosin-Deficient Congenital Muscular Dystrophy with Occipital Lissencephaly

Mustafa Mert Aydın, Munis Dündar, Mehmet Canpolat, Hüseyin Per, Ayten Güleç

3rd Place

A Rare Case: Smith-Magenis Syndrome

Ziya Bulduk, Momen Kanjee, Büşra Saruhan, Dilara Aydemir, Çiğdem Yüce Kahraman, Abdulgani Tatar

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Abstracts / Özetler

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8th INTERNATIONAL CONGRESS OF MEDICAL GENETICS
September 21-23th 2023
Invited Speakers

ceRNA Network in Neurodegenerative Disorders

Yunus Kasım Terzi

Başkent University Faculty of Medicine, Department of Medical Genetics, Ankara, Türkiye

Abstract

Neurodegenerative diseases are a group of disorders characterized by the progressive degeneration of the structure and function of the nervous system. Although, we know that oxidative stress, mitochondrial dysfunction, excitotoxic amino acids, and inflammations are major pathogenesis involved, the causes of neurodegenerative diseases remain unknown. The competing endogenous RNA (ceRNA) network is a regulatory network that involves various RNA molecules, including messenger RNAs, long non-coding RNAs, and microRNAs (miRNAs). According to the ceRNA hypothesis, RNA-RNA cross-talk via miRNA response elements might build a large-scale regulatory network spanning the transcriptome that includes both coding and non-coding RNAs (ncRNAs). Many genes implicated in neurodegenerative diseases are part of the ceRNA network. By influencing the expression of these genes, ceRNA interactions can impact disease progression. The ceRNA network is highly complex, with multiple interactions occurring simultaneously. This complexity makes it challenging to decipher the precise role of specific ceRNA interactions in neurodegenerative diseases. It is necessary to use computational methods and experimental validation to identify and study relevant ceRNA interactions. Understanding the ceRNA network's role in neurodegenerative diseases may offer potential therapeutic targets. Modulating ceRNA interactions could be a strategy to restore normal gene expression patterns and potentially slow down or halt disease progression. The ceRNA network is an intriguing concept in the context of neurodegenerative diseases. It highlights the intricate interplay among different RNA molecules and their potential contribution to disease pathogenesis. While research in this area is ongoing, it holds promise for uncovering new insights into disease mechanisms and developing novel therapeutic approaches.

Genetics of Hypokinetic Diseases and Genetic Counseling

Zerrin Yılmaz Çelik

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Abstract

Hypokinetic diseases, a sub-branch of movement disorders, include a group of neurological conditions characterized by reduced voluntary movements. These diseases usually develop after a genetic disorder. These disorders can be transmitted from the family or may occur uniquely to the individual. A family history of hypokinetic disease may be a strong indicator of a genetic component. Individuals whose close relatives have been diagnosed with these diseases may have a higher risk due to shared genetic factors. Advances in genetic research have led to the investigation of gene therapy as a potential treatment for some hypokinetic diseases. Genetic information can enable the practice of precision medicine by tailoring treatment plans to an individual's genetic makeup. Current genetic diagnostic tests can help identify specific mutations or genetic variations associated with certain hypokinetic diseases. This can provide valuable information for both diagnosis and treatment planning. It is important to note that genetic disorders alone do not determine the development of hypokinetic diseases. To understand and manage these complex neurological disorders, it is necessary to consider both genetic and environmental factors. Environmental factors, such as exposure to toxins or lifestyle choices, can also play an important role in the onset and progression of diseases. Genetic counseling is important in terms of assessing genetic risk after family tree analysis, discussing potential consequences, and providing guidance for patients to make informed decisions about themselves. As a result, genetic approach to hypokinetic diseases; by examining the role of genetics in the pathogenesis of the disease, it aims to identify the relevant genetic factors, understand the personal/familial genetic and clinical risks of these factors and contribute to their management. In this presentation, it is aimed to discuss genetic approach and counseling by providing updated information about hypokinetic diseases and related genetic causes.

Epilepsies and Current Treatment Approaches from the Perspective of a Pediatric Neurologist

Rabia Tütüncü Toker

Bursa Uludağ University Faculty of Medicine, Department of Pediatrics, Division of Neurology, Bursa, Türkiye

Abstract

Although epilepsy is a historical disease, with the rapid progress of studies in the field of genetics, there is a need for new updates in terms of both definition, classification and treatment. The International League Against Epilepsy defined epilepsy as at least 2 untriggered seizures recurring at intervals longer than 24 hours; a diagnosis of epileptic syndrome, or one untriggered seizure accompanied by >60% probability of recurrence. Epilepsy; affects 5-10 per thousand of children, with the highest incidence in infancy. This rate varies between 8-14 per thousand in Türkiye. Epilepsy syndromes are defined and classified in newborns, infants and children in 2022 by considering certain criteria such as age of onset, family history, neurological examination, possibility of responding to medication, remission, comorbidities, seizure types, electroencephalography features, neuroimaging, genetics, and differential diagnosis. Epilepsy syndromes in newborns and infants were classified as self-limited epilepsies, developmental and epileptic encephalopathies and etiology-specific syndromes. Childhood epilepsies were grouped as self-limiting focal epilepsies of childhood, genetic generalized epilepsies and developmental and/or epileptic encephalopathies of childhood. Knowing the type and etiology of epilepsy and confirming the clinical suspicion of a specific syndrome avoids unnecessary additional tests. Contraindicated medications and ineffective treatment are avoided. Such as avoiding sodium channel blockers in patients with SCN1A-related epilepsy, or preferring these drugs first in channelopathy caused by KCNQ1 mutation in the neonatal period. It allows predicting the prognosis and allowing for information. It helps to personalize treatment based on the etiological gene.

Keywords: Epilepsy, epilepsy syndromes, gene, treatment

The Use of Systems Biology in Treatment of Liver Diseases

Adil Mardinoğlu

Science for Life Laboratory, KTH - Royal Institute of Technology, Stockholm, Sweden; Centre for Host-Microbiome Interactions, Faculty of Dentistry, Oral & Craniofacial Sciences, King's College London, London, United Kingdom

Abstract

To develop novel strategies for prevention and treatment as well as to gain detailed insights about the underlying molecular mechanisms of liver diseases, it is vital to study the biological functions of liver and its interactions with other tissues and gut microbiota. Biological networks can provide a scaffold for studying biological pathways operating in the liver in connection with disease development in a systematic manner. In my presentation, I presented our recent work, where biological networks have been employed to identify the reprogramming in liver physiology in response to fatty liver disease. Based on this mechanistic modelling approach, we identified novel drug targets which may lead to design of targeted and effective treatment for liver diseases. I also presented how we developed number of drug candidates and showed their effect by performing *in vitro* and *in vivo* studies. Our study is a great example for discovery of novel drug targets and development of drug candidates for modulating our drug target.

Decoding Diseases in 3D: AI-powered Cell Level Imaging and Omics

Ali Ertürk

German Research Center for Environmental Health (GMBH), Helmholtz Zentrum München, Germany

Abstract

Our research integrates cell-level imaging by tissue clearing and omics methods via deep learning to accelerate drug development. We focus on visualizing complex biological systems at the single-cell level, including whole mouse bodies and centimeter-size human tissues. We combine this 3D-imaging data with proteomics to characterize pathologies and therapeutic effects including toxicity and efficacy. Unbiased 3D cell-level imaging on complex biological systems leads to quicker discoveries for diseases such as neurodegeneration, cancer, and diabetes. Exemplary applications of cell level assessment in whole mouse body, organoids, and large monkey and human tissues.

Modeling Neurodevelopmental Disorders in *Drosophila melanogaster*

Arzu Çelik Fuss

Boğaziçi University Faculty of Medicine, Department of Molecular Biology and Genetics, İstanbul, Türkiye

Abstract

Drosophila melanogaster (fruit fly) is a powerful model organism in the study of neurodevelopmental disorders (NDDs). NDDs have a global impact, affecting over 15% of children and covering a broad spectrum of early-onset syndromes that alter the development of the central nervous system. The severity of these disorders ranges widely and includes conditions such as intellectual disability, developmental delay, and autism spectrum disorder, among others. The mechanisms leading to NDD involve nervous system development, ubiquitination, chromatin modification and transcription regulation, signal transduction, synaptic function, metabolism, cytoskeletal dynamics, and Rho GTPase signaling. My presentation will focus on the *RNFT2* gene, which has not yet been linked to any disease. A missense mutation in RNFT2 (c.1150T>C, resulting in p.C384R) was investigated for its role in intellectual disability. This mutation affects zinc ion binding and alters protein stability. We show that the *Drosophila* homolog of RNFT2, CG13605, is expressed in the mushroom bodies, which are crucial for learning and memory. Our study utilized CRISPR/Cas9 technology to create CG13605 mutants, revealing significant morphological problems in mushroom body structure and a shorter lifespan. Negative geotaxis and courtship behavior tests were conducted to assess the mutants' behavioral aspects. Interestingly, human RNFT2 was shown to rescue the defects in CG13605 mutants, suggesting functional conservation and potential therapeutic avenues. This research demonstrates the efficacy of *Drosophila* as a model for understanding the genetic and molecular underpinnings of neurodevelopmental disorders, paving the way for future medical genomics applications in diagnosing and treating these conditions.

Keywords: *Drosophila melanogaster*, neurodevelopmental disorders, RNFT2

Genetics of Epilepsy and Genetic Counseling

Aslıhan Kiraz

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Abstract

Epilepsy, a neurological disorder characterized by recurrent seizures, manifests as a heterogeneous group of conditions, with genetic factors emerging as pivotal contributors to its pathogenesis. Various genes have been implicated in elevating susceptibility to epilepsy, offering crucial insights into the disorder's underlying mechanisms. These genes, influencing key aspects of brain function such as ion channel regulation, neurotransmitter release, and neuronal excitability, when mutated, disrupt the delicate balance of neuronal activity, culminating in abnormal electrical discharges and seizure manifestation. Genetic testing assumes a crucial role in the diagnostic process, facilitating the identification of causative mutations. As our comprehension of the genetic architecture of epilepsy evolves, genetic testing becomes increasingly integral in informing clinical decision-making. Treatment modalities for genetic epilepsy encompass a spectrum including antiepileptic drugs, ketogenic diets, and, in certain cases, surgical interventions. Ongoing research endeavors focus on unraveling intricate genetic interactions contributing to epilepsy, holding promise for novel therapeutic targets. Within the comprehensive management of epilepsy, genetic counseling provides indispensable information and support to affected individuals and their families. Precision medicine, leveraging unique genetic profiles, holds potential for more individualized treatment strategies. Advancements in gene-editing technologies further offer prospects for targeted correction of pathogenic mutations, introducing a potential curative dimension to the management of genetic epilepsy. In sum, the integration of genetic insights into diagnosis, treatment, and counseling underscores a transformative paradigm in addressing the complexities of epilepsy.

Keywords: Epilepsy, genetic counseling, neurogenetic disease

Identifying the Route from Mutation to Function from the Perspective of Elucidating Neurodevelopmental Disorders

Gülden Budak

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Abstract

Neurodevelopmental disorders (NDDs) with a heterogeneous etiology are characterized by impairment in cognition, communication, adaptive behavior and psychomotor skills. NDDs include autism spectrum disorder, impaired cognition and epilepsy. Considering that the diagnosis period of single gene disorders varies between 5-30 years, molecular basis of gene-disease relations needed to be determined. Microcephalic Primordial Dwarfism (MPD) is a group of rare diseases consists of four different subgroups of syndromes named Seckel syndrome, Meier-Gorlin syndrome, Microcephalic Osteodysplastic Primordial Dwarfism Type II (MOPDII) and Type I/III (MOPDI/III). Common characteristics of MPD include severe prenatal and postnatal growth retardation, skeletal dysplasias, agenesis of the corpus callosum, impaired development of the cerebral cortex (pachygyria/agiri), microcephaly, ıntellectual disabilitiy and epileptic seizures. We evaluated three different family affected from Renpenning syndrome, Seckel syndrome and MOPDI/III. In all three families, genome-wide homozygosity mapping and exome/DNA sequencing analyses identified the genes responsible for the disease. We identified *PLK4* as a new *Candidate* gene in the patient affected by Seckel syndrome and demonstrated by functional studies that it plays a role in response to DNA damage. Third family was linked to MPD, where we identified a gene of unknown function by genome-wide homozygosity mapping-exome sequencing. In this context, we cloned the *Candidate* gene and delivered it into HEK293 cell line by stable transfection. We identified its interaction partners by Immunoprecipitation and Mass Spectrometry analysis. Immunofluorescence staining was also used for identification of cellular localization of protein of interest which may partly give information about its cellular processes.

Keywords: *Candidate* gene identification, DNA sequencing, genetic mapping, protein-protein interaction

Comparative Analysis of Familial Exome Analysis Tools: Finding Low Frequency Rare Variants in Familial Multiple Sclerosis

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Abstract

Analyzing familial exome data involves various steps, can be complex, requiring a combination of different tools to process and interpret the genetic information effectively. Among the publicly available tools like Exomiser, famFHAT, and pVAAST, Exomiser focuses on phenotype associations, while famFHAT integrates pedigrees and covariates such as severity scores. However, linear models bypass gene functionality and pathway nuances, whereas knowledge-based methods might miss genetic variations. We employed linear methods famFHAT, pVAAST, burden tests and relatedness enriched RandomForest model, alongside knowledge graph-based techniques Exomiser and g:Profiler for enrichment analysis in Turkish Familial MS cohort consisting of 45 multiply affected families. Exomiser's top 50 genes for each family were integrated based on interfamily sharing and ranked by subgraph occurrence. Known MS-associated genes in the top 100 rankings were assessed for validation. Also, segregation pattern of variants in families was investigated with the full-likelihood bayes factor algorithm. In the famFHAT analysis of the top 100 genes, five were linked to MS with p-values ranging from 0.000450 to 0.004880, while the pVAAST analysis revealed four genes associated with MS, with p-values between 0.0 and 0.04. Likewise, the Exomiser analysis identified four MS-associated genes with p-values ranging from 0.0001 to 0.01. Functional enrichment analysis focusing on blood-brain barrier, cell adhesion and immune pathways, multiple pathogenic rare variations found in genes such as *LAMA5*, *LAMB4*, *LAMB1*, *LAMA2*, *LAMA1*, *TJP1*, *OCLN*, and *TNFRSF21* among affected individuals. Our findings provide a novel perspective on the genetic landscape of familial MS. Project was supported by GeleceğiMSin Project (2022).

Keywords: Multiple sclerosis, exome analysis, laminin genes

Genetic Basis of Hydrocephalus and Modeling Congenital Hydrocephalus Genes in the Xenopus Model System

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Abstract

Congenital hydrocephalus (CH), the pathological expansion of the cerebral ventricles due to cerebrospinal fluid (CSF) accumulation, is a common birth defect affecting 1 in every 700 births. Unfortunately, the genetic basis of CH impedes our understanding of CH pathogenesis. Although sequencing technologies enable *CH* gene discovery, we do not have a tractable animal model to test the impact of gene dysfunction on CSF circulation and CH pathogenesis. For this purpose, we developed the frog *Xenopus* as a rapid animal model to study CH pathogenesis. *Xenopus* has many advantages. Hundreds of synchronized embryos can be manipulated from the cleavage stages after in vitro fertilization. In addition, *CRISPR/CAS9* gene editing technology works well in *Xenopus,* producing biallelic gene modifications in F0 embryos, enabling rapid modeling of disease phenotypes in F0 embryos. Specifically for studying hydrocephalus, the *Xenopus* tadpole brain is semi-transparent, enabling optical coherence tomography imaging to detect 3D CSF flow throughout the entire ventricular system in living tadpoles. Using this system, we showed the visualization of a whole ventricular CSF flow network, enabling us to analyze better the interactions between the candidate human *CH* genes, ventricular development, and brain patterning. A currently held view is that CH forms due to excessive fluid/pressure in the ventricles due to obstruction or loss of CSF flow, requiring neurosurgical shunting to relieve the accumulation of this fluid. However, when we modeled candidate human genes in *Xenopus*, we showed earlier developmental defects leading to late hydrocephalus phenotype, including changes in neural cell fate.

Keywords: Hydrocephalus, *Xenopus*, optical coherence tomography

Genetic and Phenotypic Analysis of Patients with Mucopolysaccharidosis type IIIB Co-morbid with Autism Spectrum Disorder

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Abstract

Mucopolysaccharidosis type IIIB (MPS IIIB) is an autosomal recessive lysosomal disorder caused by mutations in the α-N-acetylglucosaminidase (*NAGLU*) gene. Our WES and standard Sanger sequencing effort on 220 consanguineous families, in children diagnosed with ASD, identified two recurrent damaging biallelic p.D312N and p.R234G variants in the *NAGLU* gene in seven cases of four families. All affected individuals' enzymatic assay in leukocytes clearly showed that α-N-acetylglucosaminidase was completely inactive. Structure modeling of these mutations suggested that each mutation affects the stability of the enzyme and results in a loss of activity. knn-DREMI analysis of scRNAseq data of the developing human brain identified that several genes implicated in neurodegenerative disorders are positively regulated with *NAGLU* expression. Among these genes, mutations in *CLN5* and *ZBTB20* were linked to neurodevelopmental disorders including autism. Our findings suggest that molecular and cellular mechanisms controlled by the genes positively regulated with *NAGLU* expression have promise to develop the potential treatment for neurodevelopment and neurodegeneration in patients with MPS IIIB and autism.

Keywords: NAGLU, MPS IIIB, autism spectrum disorder, lysosomal enzyme, α-N-acetylglucosaminidase, scRNAseq

Autism Spectrum Disorder and Genetic Counseling

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Abstract

Neurodevelopmental disorders are characterized by developmental deficits in cognition, language, behavior, and/or motor skills that result in impairments in personal, social, academic, and/or occupational functioning. Among chronic diseases in the pediatric age period (between 3-17 years of age), the most common reason for applying to primary healthcare services is neurodevelopmental disorders. Autism spectrum disorder (ASD) is one of them. Copy number variations (CNVs) are recommended as the first-line genetic test for ASD in guidelines. When the data of 11 studies examining CNVs are examined, it has a diagnostic rate of 1.5-20.5% (average 8.1%) in the diagnosis of ASD. Among these CNVs, variants of uncertain significance are most challenging conditions. In this case, it should be evaluated whether the detected variant relevantly associated with phenotype, whether there are previously reported cases involving a similar region, whether the breakpoints match, whether it is *de novo*/familial, whether it is related to SNVs in the same gene. In a meta-analysis study investigating the Exome Sequencing method, which included ASD cases, the diagnosis rate was found to be 8-26% (average 15%). As a conclusion genetic tests and genetic counseling providing information about the risk of other individuals in the family, risk of recurrence, preimplantation genetic diagnosis and prenatal diagnosis is important for family planning.

Keywords: Autism spectrum disorder, genetics, counselling

When should We do which Test?

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Abstract

Genetic diagnostic and screening tests are used to analyze an individual's genetic material to detect genetic changes that may cause diseases or to identify potential genetic diseases. Genetic testing is usually considered when there is a family history of genetic diseases or problems with the individual's own health. These tests are also used to screen for common diseases in the populations. The main criteria for test selection can be summarized as the purpose of the test, the type of genetic variation causing the genetic disease, age of onset and hereditary or acquired status. These tests show a wide range of methodological diversity, from chromosome analysis to whole genome analysis, and have different coverage areas. Since the sensitivity of the methods and the size and nature of the variations detected vary, the etiology of the diseases must be taken into account when selecting the tests. For this, clinical prediagnosis/diagnosis should be considered as a requirement for test selection (forwards genetics). Sometimes, in cases that cannot be diagnosed clinically, a diagnosis can be made by utilizing laboratory diagnosis (reverse genetics), but for this, the clinical evaluation of the patient should be performed before the analysis. The appropriate timing of genetic testing depends on specific periods in an individual's life. For example, pre-marital or preconception screening/diagnostic tests may be followed by preimplantation genetic diagnosis and/or prenatal diagnosis, if necessary. In the postnatal period; genetic testing for a genetic disease can be done at any stage of life, from newborn to adulthood.

Keyword: Genetic testing

Autism Spectrum Disorders from a Psychiatrist's Perspective

Gül Ünsel Bolat

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Abstract

Autism symptoms can range from mild to severe forms. Since the symptoms vary from case to case, the term autism spectrum disorder (ASD) was included diagnostic manuals. This term includes Autism, Asperger Disorder and Atypical Autism. Family studies show that after the older child is diagnosed with ASD, 7-20% of subsequent children have ASD. This prevalence increases in children with two older siblings with ASD. ASD has a important health burden on families and population. With early diagnosis, problems in the developmental process can be prevented. Patients' communication skills can be improved. Awareness of the increasing prevalence of ASD; It is important to direct the interventions of society and decision makers. Genetic studies are extremely important in terms of better understanding of this disease, follow-up of cases and family planning.

Keywords: Autism spectrum disorder, child and adolescent psychiatry, follow-up, neurodevelopmental disorder

What does a Geneticist Expect from a Neurologist?

Elif Yılmaz Güleç

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Abstract

Genetic evaluation and genetic tests play an important role in the diagnosis, classification, follow-up and treatment of neurological diseases. For this reason, a geneticist is involved in the evaluation of the patient and selection of appropriate tests beside the neurologist. In the diagnostic process, the geneticist needs to see and evaluate the patient to obtain detailed information about the patient and his/her family, to draw a pedigree and to have information about neurological findings and other system findings and complaints. In addition, it is important to get information from the patient's neurologist about the prediagnosis and differential diagnoses. In this way, it will be determined which test will be ordered first in the test ordering algorithm of neurologic diseases, which are a genetically very heterogeneous group. In this way, the diagnosis process of diseases with specific inheritance patterns will not be prolonged and financial losses will be prevented. In fact, in order to accelerate this process, it would be very efficient for neurology and medical genetics clinics to hold regular meetings and evaluate patients together.

Genetics and Genetic Counseling of Hyperkinetic Diseases

Özlem Sezer

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Abstract

Hyperkinetic movement disorders are neurological syndromes characterized by excessive movement in voluntary and automatic movements without loss of strength or spasticity. It occurs as a result of the basal ganglia being affected. The most common types are chorea, ballismus, dystonia, athetosis, myoclonus, tic, and tremor. It is divided into many subgroups. It is a heterogeneous group of diseases, each with a different pathophysiology, etiology, diagnosis, and treatment. Clinical findings are variable. Genetic counseling is also different for each individual. It is named according to the nature of the abnormal or involuntary movement that dominates the clinical picture. It is crucial to diagnose movement disorders to give accurate info and advice to patients and their at-risk relatives about genetic causes, consequences, risks, precautions, and treatment options. Early identification of at-risk individuals is vital for patients who can benefit from effective medical intervention. Predictive genetic testing is inappropriate for adult-onset conditions or asymptomatic young children at risk but should be considered if clinical findings are present. The best time to identify genetic risks and evaluate the availability of prenatal or preimplantation genetic testing is before pregnancy. Families with a known disease may consider preimplantation genetic testing an option. Due to the possibility of preventable causes in the etiology, taking preventive measures will reduce the incidence of these diseases. The best way to deal with a disease is to prevent it from occurring. The possibility of effective treatment and protection of future generations depends on accurate and early diagnosis.

Keywords: Genetics, genetic counseling, hyperkinetic, movement disorders

Functional Analysis of Autism Genes in Zebrafish Identifies Convergence in Dopaminergic and Neuroimmune Pathways and Pharmacological Candidates

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Abstract

A central challenge in the genetics of autism spectrum disorders (ASDs) is advancing from gene discovery to the identification of actionable biological mechanisms. To investigate the function of ten genes that are strongly associated with ASD in the developing vertebrate brain, we performed behavioral, whole-brain structural, circuit, cellular and molecular analyses in zebrafish mutants of ASD genes. First, we identified both unique and overlapping effects of gene loss of function on basic sensory processing and arousal behaviors. Next, we identified the forebrain as the most significant contributor to brain size differences, while brain regions involved in sensory motor control, particularly dopaminergic regions, are associated with altered baseline brain activity. Third, using whole-brain RNA sequencing in zebrafish mutants of *SCN2A* and *DYRK1A*, we identified conservation of dysregulated pathways associated with *ASD* gene loss of function in zebrafish and mammals. Using hypothesis-driven gene set

enrichment analysis, we found that dopaminergic genes are significantly enriched among downregulated genes in both mutants, while microglial genes are significantly enriched among upregulated genes. Finally, we show that both mutants display a significant increase in microglia throughout the brain, with *DYRK1A* mutants exhibiting a nearly two-fold increase in microglia. Therefore, our study implicates neuroimmune dysfunction as an important pathway downstream of select ASD genes. As a next step, we are using pharmaco-behavioral profiling to identify pharmacological suppressors of mutant behavioral phenotypes. Together, this study demonstrates the power of *in vivo* functional analyses in zebrafish to identify biologically relevant pathways downstream of ASD genes.

Keywords: Autism spectrum disorder, genetics, zebrafish, dopamine, microglia

SMA Genetics and Genetic Counseling

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Abstract

Spinal muscular atrhopy (SMA) is a rare autosomal recessive neuromuscular, where one out of each 10,000 live births can be affected by this syndrome. The atrophy is caused by the gradual loss of alpha motor neurons, either within the ventral spinal cord or motor nuclei within the lower brainstem. In this study, we aimed to evaluate the carrier frequency of *SMN1* mutation causing SMA in Turkish and Turkish Cypriot populations. This is the first study to evaluate the *SMN1* deletion mutations in Turkish Cypriot population. We compare our results with the Turkish Ministry of Health and will be presented at the conference. Our findings revealed that the carrier frequency of mutation in the *SMN1* gene for exon 7 is 2% and exon 8 is 2% in Turkish population and 4% in Turkish Cypriot for both exon 7 and exon 8 deletions. In conclusion, health precautions must be taken due to the high frequency of SMA linked to the deletion of the *SMN1* gene. Carrier testing as a technique for genetic counselling may be advantageous for individuals with a positive family history. To this population, we strongly recommend premarital testing.

Keywords: SMA, genetic counselling, carrier status, exon 7, exon 8

Clinical Aspects of Multiple Sclerosis

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Abstract

Multiple sclerosis (MS) is the second most disabling disease of young adults in their reproductive ages after accidents. In my talk, "Clinical Aspects of MS", I will try to concentrate on the changes in the terminology of clinical phenotypes of MS and also I will address the relation of clinical and genetical findings. Classically 4 types of MS were defined in 1996: Relapsing-remitting (RR) (85% of all MS patients), secondary progressive (SP), primary progressive (10-20% of all MS patients), and progressive relapsing. If left untreated, most of the patients with RRMS are expected to become SP at the end of 19 years. Clinical isolated syndrome, RRMS, PPMS, SPMS were the accepted clinical MS phenotypes according to the 2013 revision. MS may be claasifed according to age of onset of MS as childhood (<18 year), adult (18-50 years), late adult (>50 years) or very late onset (>60 years). During relapsing phase, clinical attacks or radiological new forming or gadolinium enhancing lesions serve as markers of MS. In progressive phase, disability worsens in dependent of attacks. Vision loss, diplopia, defects in visual field, sensory findings (most frequent initial finding), motor deficits, cerebellar/brain stem/spinal cord/bowel and urinary system/sexual/cognitive dysfunctions, neuropsyhiatric sypmtoms, seizures, sleep difficulties, pain, MS-associated fatigue, extrapyramidal findings or paroxysmal symptoms like Lhermitte's sign, Uhfhoff's phenomenon may be present alone or in different combinations as MS suggesting signs and symptoms.

Keywords: Multiple sclerosis, clinical phenotypes, relapsing, progressive

Hyperkinetic Movement Disorders and Current Treatments from the Perspective of Neurologist

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Abstract

The term movement disorders (MD) basically describes neurological conditions characterized by excessive or excessive movement in voluntary and automatic movements without loss of strength or spasticity. Lack of movement is defined as hypokinesia (decrease in movement amplitude), bradykinesia (slowing of movement) and akinesia (loss of movement). Excessive movement is expressed as hyperkinesis (increased movements) and dyskinesia (abnormal movements) or "abnormal involuntary movements". The main hyperkinetic MDs are: dystonia, tremor, chorea and myoclonus. There are significant difficulties with the clinical diagnosis of dystonia. Great progress has been made in recent years regarding its etiological basis. It is possible to reach the correct diagnosis with the differential diagnosis list created after a good clinical evaluation and genetic analysis performed by an experienced team-laboratory. Symptomatic treatment of focal dystonia; botulinum neurotoxin, oral medications and rehabilitation. Therapeutic indications and potential stimulation targets for deep brain stimulation (DBS) continue to expand. Pathogenesis-based treatments, including gene therapy, may be possible in the near future. Tremor is a common hyperkinetic MD. Oral medication options are limited and also its systemic side effects could be seen. DBS and MRg-focused ultrasound are effective treatment methods forr tremor. After careful patient selection by the MD specialist neurologist, then the appropriate DBS target is selected. Many studies continue to be carried out regarding the treatment of chorea, especially Huntington's disease, with an increasing number in recent years. Myoclonus is seen with many etiological causes and a heterogeneous clinical picture. Therefore, treatment studies that include only patients with isolated myoclonus are limited. There is increasing evidence that DBS is particularly effective in DYT 11 myoclonus dystonia.

The Molecular Landscape of ALS in Türkiye

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Abstract

Amyotrophic Lateral Sclerosis (ALS) is a neurological disease characterized by the degeneration and subsequent loss of the upper and lower motor neurons. The list of ALS genes continues to expand, with up to 20% of ALS heritability linked to genetic variants. In addition to these genetic associations, cumulative environmental exposures and epigenetic modifications appear likely to promote an individual across the disease-onset threshold. Thus, neurodegeneration in ALS reflects a complex interplay between genetic factors and the environment, with consequent dysfunction of molecular pathways and network circuitry. Here we present our results over a cohort of 2320 ALS patients, consisting of 501 fALS and 1819 sALS cases. The four major ALS genes C9ORF72, SOD1, TARDBP and FUS solved 35% of familial and 6% of apparently sporadic cases in our cohort. Further investigation of the remaining samples with Whole Exome Analysis increased the diagnostic rate to 45% in fALS and to 10% in sALS. Understanding the genetic contributions to disease is crucial in illuminating the major molecular pathways disrupted in ALS. Most importantly, under the surface of the seemingly disparate clinical, syndromic and diagnostic classification, not only shared genes and phenotypes, but also common mechanisms and pathways may be hidden. In the new era of translational medicine, diagnoses and potential therapies are based on genetic mutations and genetic screening, and counselling gain significance.

Keywords: Amyotrophic lateral sclerosis, ALS, genetic counselling, neurogenetics, genetic screening

Neuromuscular Diseases from a Pathologist's Perspective

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Abstract

The accurate histopathological evaluation of neuromuscular diseases requires a robust clinico-pathological collaboration. Neuromuscular diseases involve a wide range of healthcare professionals, starting from those dealing with newborns in delivery rooms to various specialists at different levels of expertise. The histopathological assessment of these diseases largely relies on morphological findings, with only a small portion of these findings being disease-specific. To achieve the best evaluation, pathology laboratories specific to these diseases, rather than general pathology

laboratories where all procedures are performed, should be established. The most crucial sources of data in neuromuscular diseases are biopsies of striated muscle and peripheral nerves. The preferred method for muscle biopsy is incisional biopsy, while needle biopsies can be used in compulsory situations. Biopsies must be evaluated alongside sufficient and appropriate clinical and laboratory data. The accurate selection of the muscle to be biopsied and the biopsy site is of utmost importance. In ideal conditions, hematoxylin and eosin stains, histochemical examinations, enzyme histochemistry, and, when necessary, immunohistochemical examinations are applied to muscle biopsies in the laboratory. Electron microscopic evaluation is required for the diagnosis of some diseases. Biopsies should be of a quantity and quality sufficient for these examinations. Nerve biopsy is a less commonly used histopathological examination method due to the necessity of sacrificing the nerve being biopsied. Peripheral nerve biopsy may be considered in peripheral neuropathies and certain muscle diseases. Sural nerve biopsy is most commonly performed due to the inability of the biopsied nerve to function. Motor nerves are not subjected to biopsy. To obtain a healthy result, the physician who examines the patient should collaborate with the pathologist before performing the biopsy and should continue this collaboration throughout the biopsy examination process.

Keywords: Neuromuscular disease, histopathology

Genetics and Genetic Counseling in Spinocerebellar Ataxias

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Abstract

Spinocerebellar ataxias (SCA) are a group of neurodegenerative diseases occurring usually at the age of 30-40 years or older with a slow progression and autosomal dominant inheritance. In almost all patients, cerebellar atrophy accompanies to gait ataxia, dysarthria, and visual impairment. More than 50 loci and genes have been identified (https://neuromuscular.wustl.edu/ataxia/domatax.html). Disease-causing mutations are often the expansion of tandem repeats within the disease gene and are most commonly seen as polyglutamine (CAG) defects. Other mutations are point mutations, deletions, insertions, duplications in related genes. Disease mechanisms of SCAs include gain of toxic RNA function, mitochondrial dysfunction, channelopathies, autophagy and transcription-dysregulation. While CAG expansions responsible for SCA1, SCA2, SCA3, SCA6, SCA7, SCA17, DRPLA and SCA12, there are different expansions in SCA8, SCA10, SCA27B, SCA31, SCA36 and SCA37. It has been reported 28 genes in which are identified non-repeat mutations. Diagnostic tests for expansions sequences are fragment analysis, triplet primed polymerase chain reaction and long read sequencing. For other mutations, targeted next generation sequencing panels and whole exome sequencing are preferred. SCA, which is inherited as an autosomal dominant disorder, has a 50% risk of recurrence in the next generation. Asymptomatic individuals, can be detected in types with a known gene. Since there is no cure yet, genetic counseling should be provided with psychiatric support.

Keywords: Spinocerebellar ataxias, tandem repeat expansions

Muscular Dystrophies and Genetic Counseling

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Abstract

Muscular dystrophy (MD) is a group of inherited genetic diseases in which muscles weaken and degenerate over time as a result of hereditary deficiencies in the muscle membrane or accessory proteins. Genetic counseling is the process that includes providing information to individuals who carry or are at risk of carrying a genetic disease and their family members about the prognosis of the disease, its treatment, if there is a possibility of recurrence, as well as which tests should be performed at what periods and their results. Muscular dystrophies can be single gene diseases as well as multigenic. In order to understand the genetic etiology in this disease group that shows genetic heterogeneity, it is necessary to know the terms of locus heterogeneity and allelic heterogeneity. Duchenne muscular dystrophy and Becker muscular dystrophy are the best known muscular dystrophies which approximately 60% are caused due deletion, 30% due point mutation and 10% by duplication. Other diseases with well-known genetic etiologies are Limb Gİrdle, facioscapulohumeral and myotonic dystrophies. It is necessary to choose the diagnostic test method according to the type of mutation in the etiology and to provide genetic counseling both before and after the genetic testing.

Keywords: Muscular dystrophy, genetic counseling, heterogeneity

Genetics for All: Basic Principles, Family History, and Inheritance Patterns of Neurogenetic Diseases

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Abstract

A detailed clinical investigation is essential to assess the inheritance model of the disease afflicting a family, considering also possible clinical variability. Investigator notes any parental consanguinity and any natural abortions. We are most familiar with monogenic diseases with Mendelian inheritance, but many diseases exhibit complex inheritance, the best-known of which is decreased penetrance that can range from 0.99 to <0.01 per cent. Very low penetrance is considered susceptibility instead. Reduced penetrance as well as variable expressivity is due to a modifier variant in some other gene. Such modifiers either manifest a severer phenotype or hinder development of the disease. Mutations in a gene can lead to different diseases with no clinical overlap and different inheritance patterns, as in *TBC1D24*. Mutations in the same gene can cause a disease involving a single trait such as polydactyly or a syndrome with neurological findings when the mutation burden of the individual is high, as in Bardet-Biedl syndrome. Lastly, gonadal mosaicism is possible in diseases in single sibships, as in Duchenne muscular dystrophy. Many cases have been solved by exome sequencing of parent and sibs to detect such *de novo* mutations, which are not rare for genes responsible for dominant diseases. Almost all cases with dominant diseases that hinder reproduction in any way are due *de novo* mutations, and others due to somatic mosaicism. Mutations of mitochondrial DNA leading to severe disease are often due to gonadal mosaicism, mimicking recessive inheritance. All these phenomena need to be taken into consideration in assessing the pattern of inheritance.

Keywords: Complex inheritance, pedigree

New Generation Treatments for Epilepsis

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Abstract

Epilepsy is a disease historically associated with evil spirits and mystery. The long history of epilepsy dates back to a 4000-year-old Akkadian tablet found in Mesopotamia; It depicts a person “with his neck turned to the left, his hands and feet tense, his eyes wide open, and foam flowing from his mouth without him realizing it”. About a thousand years later, Late Babylonians wrote a diagnostic manual called Sakikku, which included texts describing epilepsy. Documentation on epilepsy also dates back to about B.C. It is also found in Chinese texts dated to 770-221. A group of physicians published the Yellow Emperor’s Classic of Internal Medicine, the Huang Di Nei Ching, which outlined generalized seizures. The spiritually based pathophysiology of epilepsy dates back to 3000 BC, when the Hippocratic School in Greece suggested that the brain might be the root cause of epilepsy. It remained largely unchallenged until the 5th century. Aristotle, an important philosopher of the 4th century BC, suggested that epilepsy and sleep arise from similar mechanisms. The Hippocratic idea that epilepsy was a brain disease finally gained traction in Europe from the 17th century onwards and continued throughout the millennium. Samuel Tissot (1728-1797), a prominent Swiss physician, published Traité de l’épilepsie in 1770. John Hughlings Jackson (1835-1911) laid the scientific foundation of epileptology and studied the localization of lesions that could cause seizures. He published the influential text “The Study of Convulsion”, which was the culmination of his scientific findings. The current focus of gene therapy strategies for epilepsy is primarily on neuropeptide Y, galanin, etc. It aims to reduce neuronal excitability through overexpression of neuromodulatory peptides, such as, or through genetic modification of astrocytes, for example, to suppress adenosine kinase expression.

Keywords: Epilepsy, antiepileptic treatment, gene therapy, seizure

Application of High-Throughput Sequencing Technologies in Development of Neurogenetic Disorders

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Abstract

NGS technology is speeding up neurogenetics research, helping to unravel the intricacies associated with it. Neurological disorders are not predictable in the early-stage and different people show different responses to treatment. This difference can be caused by molecular pathways

or environmental effects. Therefore, as a first step selecting a correct genetic test for diagnosis is important, which should be customized to fit the disorder. Microarray, single gene analysis and repeat expansion disorders could be used as rapid first-level tests to address these issues. If the associated results are negative, NGS methods such as whole exome sequencing (WES) and whole genome sequencing (WGS) can be considered as second-level testing platforms. WES is most commonly used due to its low associated cost and turnaround time, compared with WGS. However, the overall diagnosis rate remains low, primarily due to challenges associated with the detection of pathogenic mutations, which may be classified as a variant of insignificance. So interpretation of variants as a pathogenic or non-pathogenic become challenging as many variants are unknown or not reported previously. Another issue is that many variants may lie into non-coding regions which comprises 90% of the genome and they can't be captured by short sequencing read technologies. However, over the last decades third generation sequencing emerges to overcome some shortcomings of NGS. Specifically, long read technologies have the potential to identify molecular mechanisms associated with the neurological disorders that are caused by expansion repeats such as Friedreich ataxia, spinocerebellar ataxias, Alzheimer's disease.

Keywords: NGS technology, neurogenetics research, challenges

SMA Treatment Innovations

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Abstract

Spinal muscular atrophy (SMA) is an autosomal recessive genetic associated with the survival motor neuron 1 gene (SMN1), leading to reduced levels of the survival motor neuron (SMN) protein, results in the death of motor neurons in the anterior horn (apoptosis). The deficiency of SMN protein contributes to motor neuron loss and disease progression. Currently, there are three approved treatment methods for SMA: Onasemnogene abeparvovec (Zolgensma®), Nusinersen, and Risdiplam, all of which aim to increase SMN protein levels and alter the disease's course. Onasemnogene abeparvovec utilizes an adenovirus-associated vector serotype 9 to modify the human SMN1 gene, aiming to express SMN protein in motor neuron cells via a single-dose intravenous application. Nusinersen, when administered intrathecally, regulates the maturation of SMN2 mRNA, thereby increasing SMN protein synthesis in the central nervous system. Risdiplam operates similarly but shows efficacy not only in the central but also the peripheral nervous system, administered orally. Studies continue to explore combination therapies, high-dose applications, sequential treatments, and long-term efficacy and side effect profiles, with the aim of better understanding the advantages of each treatment option. Additionally, gene therapy, such as recombinant adeno-associated virus serotype 9 gene therapy, has shown promise in the treatment of SMA. The therapeutic sdevelopments for the treatment of SMA are focused on restoring SMN expression, which is crucial in addressing the underlying cause of the disease. In this presentation, the current treatment options for SMA were discussed. Case studies were presented.

Pathophysiology of Multiple Sclerosis

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Abstract

Multiple sclerosis (MS) is an autoimmune and degenerative disease of the central nervous system. Characterized by both inflammatory and neurodegenerative components, this condition is the most common non-traumatic disabling neurological disorder among young adults. Women are approximately three times more likely to be affected by this disease compared to men. The tissue damage in MS results from a complex and dynamic interplay among the immune system, glial cells (including myelin-producing oligodendrocytes and their precursors, microglia, and astrocytes), and neurons. Although the exact cause of this autoimmune response remains elusive, it is thought to involve a combination of genetic, environmental, and immunological factors. T-lymphocytes play a significant role in the immunopathogenesis of MS, operating through various T-cell subtypes. Th1 cells contribute to the autoimmune response in the central nervous system by secreting pro-inflammatory cytokines (interferon-γ and tumor necrosis factor-α). Th17 cells can initiate neuronal damage by producing interleukin-17. Regulatory T-cells (Tregs) play a crucial role in suppressing the autoimmune response, and their dysregulation is implicated in the pathogenesis of MS. B lymphocytes are also influential in MS pathogenesis through their role in antibody production. In individuals with MS, B-cells contribute to lesion development and disease progression. T-cells assist in activating B-cells and triggering their antibody production, while B-cells also influence T-cells by presenting antigens and enhancing T-cell activation. Innate immune system cells are essential in the pathogenesis of MS. Well-known environmental risk factors for the development of MS include low vitamin D levels, Epstein-Barr virus infection, and smoking. In conclusion, MS is an autoimmune-mediated demyelinating and neurodegenerative disease that emerges under the influence of environmental factors in genetically predisposed individuals. T-lymphocyte autoimmunity forms the

basis of its pathophysiology, with various immune system elements contributing to autoimmune-mediated central nervous system damage. The cause of autoimmunity is not definitively known. Reducing the effects of modifiable risk factors is of significant importance. Immunomodulatory treatments targeting multiple stages of pathophysiology are being used and developed. Personalizing pathophysiological understanding and treatment in MS care is among the future objectives of therapy.

Keywords: Multiple sclerosis, pathophysiology, autoimmunity, demyelination

ALS and New Treatments

Necdet Karlı

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Abstract

Motor neuron diseases (MND) are classified according to their clinical presentation as;

1. Amyptrophic Lateral Sclerosisi
2. Primary Lateral Sclerosis
3. Progressive Muscular Atrophy
4. Progressive Bulbar Palsy
5. Pseudobulbar Palsy
6. Monomelic Amyotrophy

Classical ALS is the most frequent MND making about 70% of all MND. It's clinical presentation comes with first and second motor neuron findings. Cramps and fasciculations, extremity weakness, bulbar signs (dysarthria, dysphagia, choking) may be the presenting symptoms or may be seen during the evolution of the disease. ALS is a multifactorial diseases based on a genetic mutation. 10% of ALS cases are familial. More than 40 genetic mutations have been associated with ALS. The most frequent ones being; C9ORF72, SOD1, FUS, TARDBP mutations. Diagnosis is based on neurologic examination and EMG. Treatment options are limited, all are slowing but neither curing nor stopping the disease. Riluzole, Edaravone, Sodium phenylbutyrate + tauroursodiol and tofersen are the choices for the treatment of ALS. Sodium phenylbutyrate + tauroursodiol and tofersen are recently approved treatment by FDA. Studies showed both were more promising, rate of slowing was higher than the previous treatments.

Inovations in Muscular Dystrophy Treatment: Hopes and Challenges

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Abstract

In this speech, it is privileged to share insights into the revolutionary advancements in muscular dystrophy (MD) treatment, a field witnessing remarkable strides in genetic therapy and personalized medicine. MDs, characterized by progressive muscle weakening, encompass a variety of genetic disorders. The most common, Duchenne muscular dystrophy (DMD), results from mutations in the dystrophin gene, essential for muscle function and stability. The recent surge in biomedical innovations has ushered in gene-based therapies, offering new hope. Gene replacement therapy, using Adeno-Associated virus vectors, introduces functional dystrophin genes into affected cells. Meanwhile, gene editing, particularly CRISPR/Cas9, promises to correct mutations at the DNA level. However, challenges in precision and safety persist. Exon skipping, another innovative approach, uses antisense oligonucleotides to bypass mutated gene sections, facilitating the production of a functional dystrophin protein. This technique has led to specific therapies like Eteplirsen for certain DMD mutations. In parallel, symptom management remains vital. Corticosteroids, the current standard, improve muscle strength, while emerging drugs target inflammation and fibrosis, key factors in MD progression. Looking ahead, the future of MD treatment lies in tailoring therapies to individual genetic profiles, enhancing effectiveness while minimizing side effects. Research continues to evolve, with combination therapies targeting multiple disease facets promising a more holistic approach. Our journey towards effective MD treatment is challenging but filled with potential. Through continued innovation and dedication, we aspire not just to treat MD but to transform the lives of those affected.

Keywords: Muscular dystrophy, gene therapy, gene editing, duchenne

Developing Dynamic Structure-based Pharmacophore and ML-based QSAR Models for the Discovery of New Anti-Cancer Therapeutics: Computational Drug Repurposing Efforts

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Abstract

RET holds paramount importance. Small molecules exhibit potential as inhibitors, binding to the kinase domain of RET and hindering its enzymatic activity. Nevertheless, the emergence of resistance, attributed to single amino acid changes, presents a formidable challenge. In this talk, a structure-based, dynamic pharmacophore-driven approach that utilizes E-pharmacophore modeling derived from molecular dynamics trajectories will be introduced. The objective is to identify hypotheses with low-energy favorability, employing machine learning-trained QSAR models to predict pIC_{50} values for compounds. To achieve this, a comprehensive screening of extensive small molecule libraries was conducted using developed ligand-based models. The outcome suggests potent compounds with the capability to inhibit RET activation.

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[OP-01]

A Case of Rubinstein-Taybi Syndrome Presenting with Novel Mutation Detected in the *CREBBP* Gene and Severe Dysphagia

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Abstract

Rubinstein-Taybi syndrome is an autosomal dominant multiple congenital anomaly syndrome characterized by a characteristic facial appearance and wide fingers and toes. Here, we present an 8-month-old case with a c.2095_2096insGG(p.Gln699Argfs*4) heterozygous novel mutation in the *CREBBP* gene and dysphagia.

Keywords: Rubinstein-Taybi syndrome, novel mutation, broad thumb, dysphagia

Introduction

Rubinstein-Taybi syndrome (RTS) is an autosomal dominant multiple congenital anomaly syndrome characterized by a characteristic facial appearance and wide fingers and toes. *De novo* mutations in the *CREBBP* gene are responsible for approximately 50-60% of the cases and in the EP300 gene for 3-8%. The etiological cause is unknown in 30% of patients (1). Here, an 8-month-old case with a novel mutation in the *CREBBP* gene is presented.

Case Report

An 8-month-old girl patient was referred to us due to dysmorphic appearance and microcephaly. Birth weight, height and head circumference were 2500 g (10p), 48 cm (10-50p) and 33 cm (10-59p), respectively. As in seen Figure 1, physical examination revealed narrow forehead, hirsutism on the forehead, mild blepharophimosis, beak nose, drooping nose tip, short philtrum, thin upper lip, mild retromicrognathia, short neck, thick fingers and toes, and genital examination was normal. With the current findings, RTS was considered in the patient. The c.2095_2096insGG(p.Gln699Argfs*4) heterozygous mutation was detected in CREBBP. This mutation is a pathogenic novel mutation that causes a frameshift and creates a premature stop codon. Parental mutation analysis results were found to be normal.

Discussion

The incidence of RTS is 1/100,000-125,000. It has been observed that patients sit up at the age of 1.5 on average, walk at the age of 2.5-3, and say their first words at the age of 2 (2). Our patient started walking at the age of 2 years and 8 months and said his first meaningful word at the age of 1.5. In two studies, the average IQ level was found to be between 36-51. Short attention span, autism-like behavior, and hyperactivity may be observed (3). Our patient's developmental tests revealed developmental delay at all stages. Strabismus, cataract, glaucoma, refractive errors, nystagmus, and retinal anomalies can be seen in older ages (4). Our patient started using glasses at the age of 4. Nutritional problems such as gastroesophageal reflux disease (GERD) are observed in approximately 68% of patients. Dysphagia is also a symptom of this disease that should be emphasized (5). Scott et al. (6) reported that a 7-year-old male patient, diagnosed at the age of 3, had a complaint of dysphagia for 2-3 years. Dilation was performed upon detection of a post-cricoid web in endoscopic examination. At the ages of 8 and 9, dilation was performed 2 more times. They evaluated this finding, which is generally associated with iron deficiency and is very rare in children, as congenital in this case diagnosed with RTS (6). Kumar and Thota (7) detected Barrett's esophagus (BE) in a 26-year-old male patient diagnosed with RTS and complaining of dysphagia. They learned from the patient's history that he had GERD for 15 years. Periodic dilatations were required during follow-up of the patient who underwent endoscopic balloon dilation. Twelve years after the diagnosis of BE, low-grade dysplasia developed in the BE segment and radiofrequency ablation was planned. Dysphagia recurred in the patient 2 years later (7). Noble et al. (8) reported 2 cases diagnosed with RTS with a history of dysphagia. The first is a 12-year-old male patient who was diagnosed in infancy. The patient, who had a history of GERD and aspiration pneumonia, had a history of dysphagia since the time of RTS diagnosis. The patient, who underwent endoscopy and dilatation many times until the age of 12, never had an esophageal biopsy. As a result of an esophageal biopsy performed at the age of 12, eosinophilic esophagitis and gastritis were detected, and his symptoms regressed with diet and corticosteroid treatment. The other case reported by Noble et al. (8) is a 10-year-old male patient with a history of dysphagia for 5 years. Following the recurrence of dysphagia after endoscopy and dilatation at the age of 9, the esophageal biopsy result performed at the age of 10 was found to be compatible with eosinophilic esophagitis. His symptoms regressed with the diet and corticosteroid treatment given (8). As seen in the literature review, it has been reported that dysphagia recurs as a result of symptomatic and interventional treatments unless the etiological cause is determined. At the age of 4, our patient was still unable to eat any food other than liquid due to dysphagia. No pathology was detected on OMD radiography. Endoscopy could not be performed because the family did not agree. At the age of 5.5, dysphagia began to regress and a follow-up decision was made.

Conclusion

Dysphagia is an important sign in RTS and it recurs as a result of symptomatic and interventional treatments unless the etiological cause is determined. This case, in which a novel mutation was detected, is presented to contribute to the literature.

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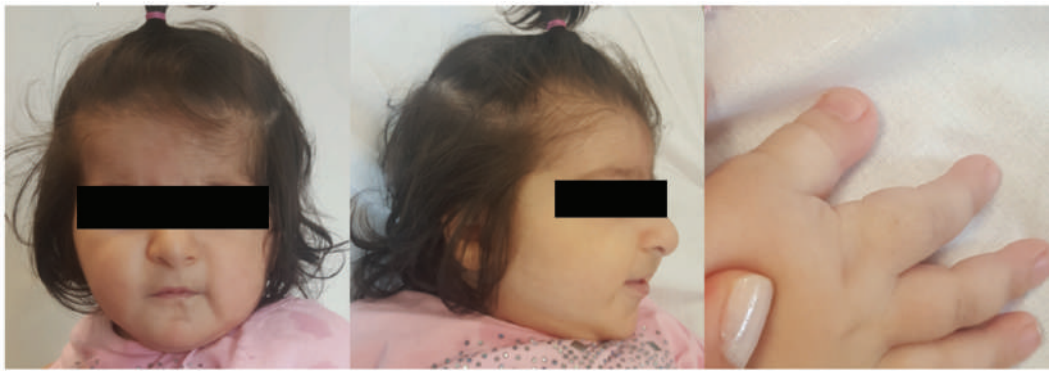

Figure 1. The patient

[OP-02]

Custom WES Panel Results in Patients with PCOS

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Abstract

Introduction: Polycystic ovary syndrome (PCOS) is a very heterogeneous endocrinopathy that can be grouped in multifactorial diseases. Studies conducted in different populations using three different diagnostic criteria have shown that its prevalence in the world is up to 20%. The potential of different variations in different genes to explain different phenotypes of PCOS has taken its place in the literature.

Methods: In our study, we evaluated 16 PCOS patients by applying androgen excess PCOS criteria and evaluated candidate genes that may be responsible for PCOS in a double-blind manner with a custom exome panel. We evaluated the patients' germline mutations and variants and whole exome analysis of 8 different genes involved in TGF-beta, WNT, MAPK, insulin signaling and steroid biosynthesis in the ION AmpliSeq Custom panel. We tried to explain the genotype-phenotype relationships of the mutations we found with clinical and biochemical laboratory data.

Results: After filtering steps, we detected 431 variations, 85 of which were different. We revealed 3 different mutations, one of which was novel, in two different patients. We described the PCOS phenotype related to the mutations we found in the *RUNX2* and *MAPK14 (p38 alpha)* genes. We have demonstrated for the first time in our population a missense variation in the *SRD5A2* gene that may affect total testosterone levels in PCOS.

Conclusion: After the segregation analyzes of the novel mutation that is located in the *RUNX2* gene and in the *MAPK14* gene are performed, it will be possible to introduce the candidate gene(s) responsible for PCOS into the literature.

Keywords: PCOS, candidate gene, exome sequencing, RUNX2, MAPK14

[OP-03]

Contributions of NGS to Transplantation Genetics

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Abstract

The development of next generation sequencing has led to a major breakthrough in the field of medical genetics. Next generation sequencing is also very important for transplantation genetics in the field of medical genetics. With the widespread use of next generation sequencing, innovations in transplantation genetics touch the lives of many patients in this field.

Keywords: Transplantation, genetics, next generation sequencing, NGS

Introduction

Transplantation is a life-saving treatment for end-stage organ failure. Organ and tissue transplantation is a vital procedure, but it can be associated with problems such as incompatibility between donor and recipient, tissue rejection and post-transplant diseases. These can be prevented by the use of advanced immunosuppressant agents and the necessary genetic testing.

Transplantation genetics is a field of research, investigation and application used in important areas such as evaluation of disease risk after organ or tissue transplantation, determination of the compatibility of donors and recipients and post-transplantation follow-up (1). Transplantation genetics, which was limited by traditional methods, has undergone a significant evolution with the development and application of next generation sequencing technology.

1. Use of Genetic Methods in the Pre-transplant Process

a) Investigation of the Factors Causing End-stage Organ Failure

End-stage organ failure is a health condition in which organs are so severely damaged that they cannot function normally. Organ failure can affect the function of important organs such as the heart, kidneys, lungs or liver. Such failures can have many causes and genetic diseases constitute an important subgroup of organ failures (2).

- Kidney Failure and Genetic Diseases: Renal failure is a condition in which the kidneys cannot fulfill their normal functions and genetic diseases can be an important cause of renal failure (2,3). For example, polycystic kidney disease, which is seen clinically after a loss of function in the *PKD1* and *PKD2* genes, leads to the formation of cystic masses in the kidneys and may cause kidney failure over time. Alport syndrome, which is associated with genes such as *COL4A3*, *COL4A4* and *COL4A5*, is associated with kidney damage, hearing loss and eye abnormalities and is usually caused by a genetic mutation (4). Fabry disease (related to *GLA* gene) is one of the lysosomal storage disorders and may lead to kidney damage (5). Such genetic diseases, which can be observed especially in pediatric patients, increase the risk of renal failure and may ultimately lead to the need for kidney transplantation (6).

- Liver Failure and Genetic Diseases: Liver failure is a condition in which the liver is damaged to the state it cannot fulfill its normal functions. For example, Wilson's disease, which is associated with the *ATP7B* gene, is an inherited disease in which copper accumulates more than normal and causes liver damage (7). Alpha-1 antitrypsin deficiency, caused by a mutation or loss of function in the *ATT* gene, is an inherited disease that causes abnormalities in the lung and liver (8). These and similar genetically based diseases may lead to liver failure by affecting the normal functions of the liver and may necessitate liver transplantation.

- Heart Failure and Genetic Diseases: Heart failure is a condition in which the heart cannot pump enough blood to the body. Cardiomyopathies are among some genetic diseases that increase the risk of heart failure. Cardiomyopathy is a genetic disease that causes abnormal growth and dysfunction of the heart muscle. Another example is the susceptibility to coronary artery disease caused by high cholesterol levels and low-density lipoprotein receptor mutations.

Genetic diseases can be one of the underlying causes of organ failure and can lead to the need for serious treatments such as organ transplantation. Early diagnosis, genetic testing and genetic counseling play an important role in preventing or managing genetic diseases and end-stage organ failure. While conventional methods investigate target genes or even some of the exons of the targeted genes, the next generation sequencing method, which has the important advantage of high data throughput, can analyze patients from a wide perspective and observe changes in multiple genes. As recently observed in our unit, multi-genic changes underlie the disease pathogenesis of many patients. For these reasons, next generation sequencing seems to be more advantageous than targeted Sanger sequencing in the diagnosis of such multigenic diseases.

b) HLA Typing and Tissue Compatibility Analysis

The HLA class I and class II loci are the most polymorphic genes in the human genome (9). Moreover, almost half of the genes in the MHC region have paralogous copies on chromosomes 1, 9 and 19, which may have enabled the rapid evolution of new functions (10). Allelic variations in *MHC* genes form the basis of histocompatibility in transplantation. Successful matching of HLA-A, HLA-B and HLA-DR is considered the most important factor. In case of HLA mismatch, the success of transplantation and the survival of the transplanted organ decrease depending on the rate of mismatch (11). Therefore, HLA typing is vital to identify mismatches and to ensure appropriate matching of donor-recipient pairs.

For effective *HLA* gene typing, many methods with low margin of error that can generate high data output have been developed (12,13). These methods, including next generation sequencing, have recently started to be applied in the field of solid-organ transplantation. The YNS method, which enables the detection of rare sequence variants in individual samples with a large number of sequence reads (300,000-400,000) in a single run, has become routinely used in the clinical setting in the last few years. This routine use is partly due to the fact that the cost of sequencing technologies has decreased over time to a level comparable to Sanger sequencing (14,15).

Even patients with HLA-compatible transplants may experience acute or chronic rejection processes, suggesting a role for non-HLA factors in alloimmunity (16). Studies have shown that graft loss in organ transplant recipients within 10 years is caused by non-HLA factors in 38% of cases, only 18% by HLA-related factors and 43% by non-HLA immunologic factors (e.g., surgical complications, drug toxicity or infections) (17,18). When non-HLA mediators are mentioned, killer cell immunoglobulin-like receptor (KIRS), MHC class 1 polypeptide-associated sequence A (MICA) and minor histocompatibility antigen (miHAs) come to mind firstly. Donor-recipient mismatches of these potential non-HLA mediators may trigger an alloimmune response through antigen-receptor interactions or polymorphic genes encoding non-specific peptides (19).

While only HLA compatibility of the patients was investigated with a single data during the transplantation process with conventional methods, the use of methods that allow obtaining multiple data such as YNS has enabled the examination of not only HLA compatibility of the patients but also immunologically important non-HLA loci whose effect on organ rejection has been scientifically proven. This allows genetic tests performed before transplantation to be maximized and have wider clinical implications due to the high data output that can be obtained.

c) Evaluation of Recipient-Donor Suitability for Transplantation by Genotype/Phenotype Relationship

In the conventional transplantation process, it is sufficient that the donor's general health condition and the condition of the kidney to be transplanted have been healthy until then and have HLA compatibility with the recipient. Nowadays, we know that some genetic diseases can occur with a late-onset manner, that is, at an advanced age. What needs to be considered especially in living transplant donors, is the possibility of a potential organ failure in the future due to genetic variations/mutations even if the donor is observed to be healthy during the pre-transplantation process. Therefore, instead of focusing only on the recipient, it is necessary to make a decision considering the health of the organ donor as well. There is a very important Latin saying in medical education: "Primum Non-Nocere", meaning "First Do No Harm". When treating our patient with organ failure, it is possible to accelerate the process of organ failure by selecting an individual as an organ donor who may have a possible organ failure in their later years as a result of genetic variations. In addition, the transplanted organ may have a late-onset genetic disease, making it highly likely that the transplanted organ will fail again in the future.

It is known that the percentage of development of pediatric organ failure on the basis of genetic factors is quite high (20). It is also quite common in Türkiye that the potential organ donors of this patient group are recipients parents. Considering these conditions, it is very likely that the mutation causing organ failure in the child is present in the parents too. Even if the carrier status of the parents for diseases with autosomal recessive inheritance pattern is a condition that requires long planning in the transplantation process, the main problem is the mutations found in genes associated with diseases with two different inheritance patterns such as COL4A3. Autosomal Recessive (MIM:203708) and Autosomal Dominant (MIM:104200) inheritance patterns associated with COL4A3 and Alport syndrome, which may progress with end-stage renal failure, have been described in the literature.

For these reasons briefly mentioned above, instead of examining only current clinical findings, genetic examination of both the recipient and the donor during the transplantation process will increase the success rate in the transplantation process. In this way, the transplanted organ will have a longer survival outcome, and the donor candidate will be prevented from becoming a transplant candidate as a result of a possible organ failure in the future.

2. Using Genetic Methods in the Post-Transplant Process

a) Rejection Monitoring

Post-transplant tissue rejection is a condition in which the recipient rejects the transplanted tissue or organ (21). Post-transplant genetic analysis can be used to provide early diagnosis of tissue rejection. Genetic material in the recipient is analyzed and immune responses can be detected. In this way, tissue rejection can be detected at an early stage and appropriate treatment can be initiated.

Analysis of circulating free DNA (cfDNA) in serum allows early diagnosis of organ rejection and increases patient comfort as it is performed with a non-invasive method (22). cfDNA in serum is DNA fragments circulating in the blood as a result of aging and death of cells. In case of organ rejection, rejection of the transplanted organ by the recipient leads to destruction of organ cells and circulation of donor-derived free circulating DNA (dd-cfDNA). Analysis of (cfDNA) in serum is based on the detection of dd-cfDNA in the bloodstream and can be performed non-invasively to help detect organ rejection at an early stage.

In rejection monitoring, blood samples are taken at regular intervals and cfDNA levels are monitored. If an abnormal increase or change in cfDNA level is observed, the risk of organ rejection is assessed and necessary measures are taken. This helps to better monitor organ survival and transplant outcome. When the tests performed in our center are examined, the destruction of the transplanted organ is recognized before it is reflected in the clinic and taking the necessary measures not only positively affects the survival of the transplanted tissue, but also contributes greatly to minimizing the loss of function.

b) Microchimerism Monitoring

Next generation sequencing is also used in microchimerism analysis. Detection of small amounts of donor cells remaining in the recipient after transplantation can increase transplantation success (23,24). High-resolution YNS facilitates the detection of microchimerism and helps to determine treatment strategies for the recipient by confirming the presence of donor cells. Especially in transplants of tissues with bone and bone marrow content such as composite tissue transplants, monitoring of microchimerisms is very important in terms of organ rejection (24).

c) Drug Level Monitoring

Next generation sequencing technology also plays an important role in pharmacogenetic analysis. Pharmacogenetics is a field that studies the effects of drugs according to an individual's genetic profile. Tacrolimus drug level monitoring is also considered in this context. Tacrolimus helps to prevent rejection of the donor organ by recipients by suppressing the immune system (25). Tacrolimus drug administered to the recipient after transplantation surgery reduces the immune response by inhibiting the activation of T-cells. This is important to ensure acceptance of the donor organ by the recipient and long-term success. Next generation sequencing analyzes the recipient's genetic profile to obtain information about their response to tacrolimus and drug metabolism. This enables more personalized and effective treatment planning for recipients.

The cytochrome P450 3A family, particularly CYP3A4 and CYP3A5, are among the best-characterized and best-studied determinants of the variability in blood levels of immunosuppressant drugs. Genetic variations in the genes coding for these cytochromes may affect the pharmacokinetics of tacrolimus and mTOR inhibitors (26,27). Many allelic variants of CYP3A4 and CCYP3A5 coding genes have been identified and shown to significantly alter the metabolism and clearance of tacrolimus, affecting the blood levels of the drug. The vast majority of these variants cause a loss of function of the enzyme and therefore significantly increase blood/serum tacrolimus levels. For example, CYP3A5*1 encodes a functional enzyme. Any mutation in this allele leads to the formation of dysfunctional proteins (28,29). For CYP3A4 in the same cytochrome family, it has also been reported in the literature that some CYP3A4*22 allelic variants cause decreased enzymatic activity (30). It should of course be noted that the frequencies of these alleles may vary widely between different population groups.

d) Prevention of Side Effects (Infection and Cancer)

By adjusting the dose of immunosuppressant agents in a patient-specific manner in line with the genetic data we will obtain individually, we have the opportunity to minimize side effects such as secondary infections and cancer caused by these agents.

It has also been shown in the literature that some genes such as *LINC00882*, *CACNA1D* and *CSMD1* and changes in these genes are associated with cancers seen after transplantation (31). Pre-evaluations of these genes have a very important place in the planning of the whole post-transplant process, starting from the selection of immunosuppressant agents to be used after transplantation.

e) Non-invasive Prenatal Diagnosis

Because of the teratogenic effects of high doses of immunosuppressive agents and some pharmacologic drugs taken during the transplantation process, the pregnancy process of transplanted individuals should be carefully monitored and some drugs should be replaced with other agents in case of pregnancy (32). Detection of fetal anomalies during pregnancy of transplant recipients is important in this respect. Therefore, prenatal diagnosis gains importance in these cases. Non-invasive diagnostic methods, which have recently gained popularity, can also be used in this patient group. Non-invasive prenatal diagnosis (NIPD) aims to detect fetal-related genetic disorders before birth, with the potential to reduce the risk of fetal birth defects by detecting markers in the peripheral blood of pregnant women.

Prenatal diagnosis by preimplantation genetic diagnosis is as important in Uterus transplants as it is in solid organ transplants. In uterine transplantation, which aims to provide prospective parents with a healthy child, it is very important to follow up the individual who has undergone uterine transplantation in terms of both maternal and infant anomalies throughout the pregnancy process. Postnatal karyotyping and NIPD tests were found to be compatible and it was shown that NIPD tests can also be applied in this patient group.

In conclusion; innovative genetic approaches increase the success of the transplantation process and positively affect the quality of life of patients. Thanks to all these scientific approaches, the use of genetic tests in the field of transplantation and the integration of next generation sequencing technology play an important role in increasing the success and efficiency of transplantation processes.

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[OP-04]

Molecular Diagnosis of Lethal Multiple Pterygium Syndrome in a Fetus Presenting with Fetal Akinesia Deformation Sequence

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Introduction

Fetal akinesia deformation sequence (FADS) is a rare disorder characterized by decreased fetal movement in utero, which leads to various joint contractures and other developmental abnormalities in the affected fetus. The genetic basis of FADS is complex and heterogeneous. Here, we present a patient presenting with FADS who has been diagnosed as lethal multiple pterygium syndrome (LMPS).

Case Report

A male patient who died in first hour following the birth was referred to pediatric genetic clinic. Hydropic appearance, flattened nasal bridge, micrognathia, low set ears, short neck, cystic hygroma, bilateral flexion contractures at elbows, fingers, hips, knees and bilateral rocker bottom feet were noticed during the postmortem examination. Antenatal ultrasonography revealed reduced fetal movement, multiple arthrogryposis, clenched hand, hydrops and pulmonary hypoplasia. His parents were consanguineous and had a history of 2 termination of pregnancy with similiar phenotype. The homozygous CHRNG:c.753_754del(p.Val253AlafsTer44) variant associated with LMPS was detected in the patient who underwent WES with a prediagnosis of FADS.

Conclusion

Mutations in the *CHRNG* gene have been reported to cause recurrent pregnancy loss and LMPS. Genetic testing, including whole genome sequencing, can help identify specific genetic mutations responsible for FADS in individual cases. However, due to the genetic heterogeneity of FADS, a precise diagnosis might not always be achievable. It's important to work closely with medical geneticists to determine the underlying cause of FADS in specific individuals. Genetic counseling is crucial to understand the inheritance pattern, recurrence risks, and potential treatment options for affected families.

[OP-05]

Imagawa-Matsumoto Syndrome: The First Case from Türkiye

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Abstract

Imagawa-Matsumoto syndrome (IMMAS; MIM #618786) is an autosomal dominant syndrome characterized by overgrowth, dysmorphic features, musculoskeletal abnormalities, developmental delay, and intellectual disability. The first case was reported in 2017 and has subsequently been diagnosed in only another 12 patients. We also present the first IMMAS patient from Türkiye. A 19-year-old female was admitted to the neurology outpatient clinic due to behavioral disorder and intellectual disability. Her physical examination revealed macrocephaly and dysmorphic features like a round face, broad forehead, hypertelorism, and variable skeletal anomalies such as flat feet, clinodactyly, and macrocephaly. Cranial magnetic resonance imaging showed agenesis of the corpus callosum and polymicrogyria. Chromosomal analysis results were consistent with a normal constitutional female karyotype and microarray analysis showed a *de novo* 1.5-MB size deletion on the long arm of chromosome 17; band q11.2 encompassing the Polycomb Repressive Complex 2 Subunit (*SUZ12* gene, MIM *606245). This report will contribute to the limited information in the literature.

Keywords: Behavioral disorder, Imagawa-Matsumoto syndrome, Intellectual disability, Macrocephaly, *SUZ12*

Introduction

Imagawa-Matsumoto syndrome (IMMAS; MIM #618786) is characterized by overgrowth, development delay, intellectual disability, and dysmorphic features. Imagawa et al. (1) were the first to report such a case with mutation in the *SUZ12* (Polycomb Repressive Complex 2 Subunit; MIM *606245) gene -an 11-year-old female (with a dizygotic twin) born to non-consanguineous Japanese parents, that had postnatal overgrowth, intellectual disability, such dysmorphic features as hypertelorism, downslanting palpebral fissures, and a prominent forehead, musculoskeletal and structural brain anomalies, in 2017. These findings were initially interpreted as Weaver syndrome (WS; MIM #277590), which is characterized by prenatal or postnatal overgrowth, accelerated osseous maturation, cranial dysmorphism, intellectual impairment, and limb anomalies; however, the patient did not fulfill all the criteria for WS. As such, the patient was diagnosed as 'Weaver-like'syndrome. Subsequently, in 2018, two new patients from unrelated families were described with postnatal overgrowth and development delay, and novel *SUZ12* mutations as Weaver-like syndrome by the same group, the clinical characteristics of these patients were very similar to the first reported patient (2). In 2019, Cyrus et al. (3) reported another 10 patients with an overgrowth phenotype, physical abnormalities, delayed developmental stages, and rare heterozygous SUZ12 variants. Considering these ten patients in addition to the previous three patients, the clinical framework of pathogenic SUZ12 variants has settled, which mostly causes overgrowth, distinctive facial features, limb anomalies, and intellectual disability. It later began to be called "Imagawa-Matsumoto syndrome".

Case Report

A 19-year-old female presented to our neurology outpatient clinic with a history of intellectual disability and behavioral disorder since childhood. The patient has non-consanguineous Turkish parents and was born via spontaneous vaginal delivery after an uneventful full-term pregnancy. She had a low birth weight (<2500 g, <3 percentile) and was hospitalized in the newborn intensive care unit for 2 months due to neonatal pneumonia. The parents reported a history of mild motor developmental delay; the patient was able to sit at age 12 months and walk at age 2 years. Notably, her speech and language skills development were delayed; her first word was spoken at age 3 years.

Upon physical examination the first remarkable feature was a dysmorphic phenotype, including a prominent forehead, round face, broad nasal ridge, and macrocephaly. The patient's head circumference was 58 cm (+2 SD), while her weight and height were within the normal range. Musculoskeletal system assessment showed flat feet, large hands, camptodactyly, clinodactyly. Her neurological examination was normal, except for cognitive impairment. In the genitourinary system examination, it was learned that she had irregular menstrual periods. She applied to the gynecologist with this symptom; subsequent suprapubic pelvic and urinary ultrasonography performed by a gynecologist revealed an ectopically located left kidney in the pelvis, which also had a rotation anomaly.

Routine blood tests were within normal limits. Cranial magnetic resonance imaging showed agenesis of the corpus callosum and polymicrogyria (Figure 1), which was followed by chromosomal analysis. Chromosomal analysis results were consistent with a normal constitutional female karyotype. Chromosomal microarray was the next step of the genetic testing algorithm and showed a copy number loss (1 copy) of 1.5 MB (195 markers) size deletion on the long arm of chromosome 17; band q11.2 encompassing the *SUZ12* gene (Figure 2). The deletion was reported *de novo* because the change in the index could not be detected in the parents.

Discussion

IMMAS is characterized by overgrowth, prominent facial dysmorphism, developmental delay, intellectual disability, and variable skeletal abnormalities. Our patient had corpus callosum agenesis and polymicrogyria; therefore, it is important to include neuroimaging in the diagnostic algorithm of this syndrome to detect structural brain abnormalities. On the other hand, our patient is the first female IMMAS patient with a genitourinary abnormality and renal anomalies, including renal ectopia which also had a rotation anomaly. In this respect, our case adds a new clinical component to the syndromic profile of IMMAS.

Conclusion

Herein we presented the first IMMAS patient from Türkiye-the 14th known case worldwide. As such, the present case report will help improve the syndromic profile of IMMAS and expand the limited IMMAS database.

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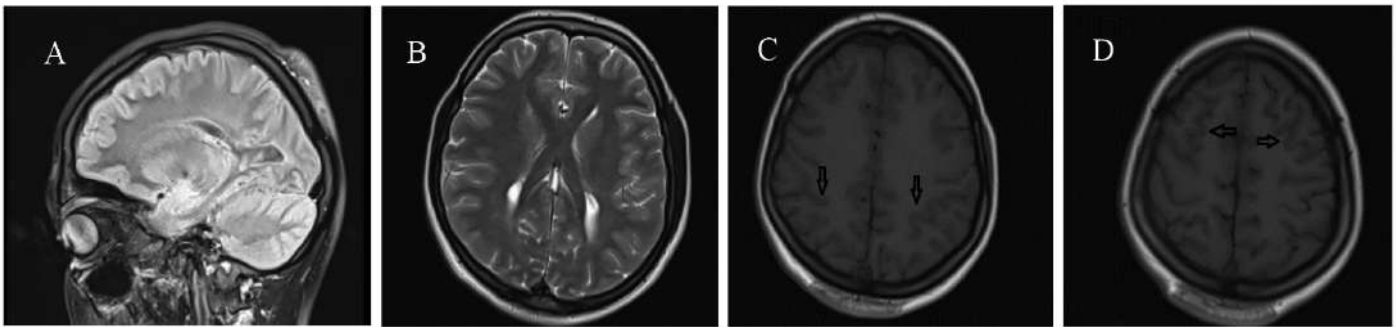

Figure 1. MRI imaging of the patient's structural brain abnormalities. MRI imaging demonstrated at sagittal (A) and axial (B) fluid attenuation inversion recovery (FLAIR) sequence agenesis of the corpus callosum and axial (C, D) T1-weighted (T1W) sequence bilaterally frontotemporoparietal polymicrogyria

MRI: Magnetic resonance imaging

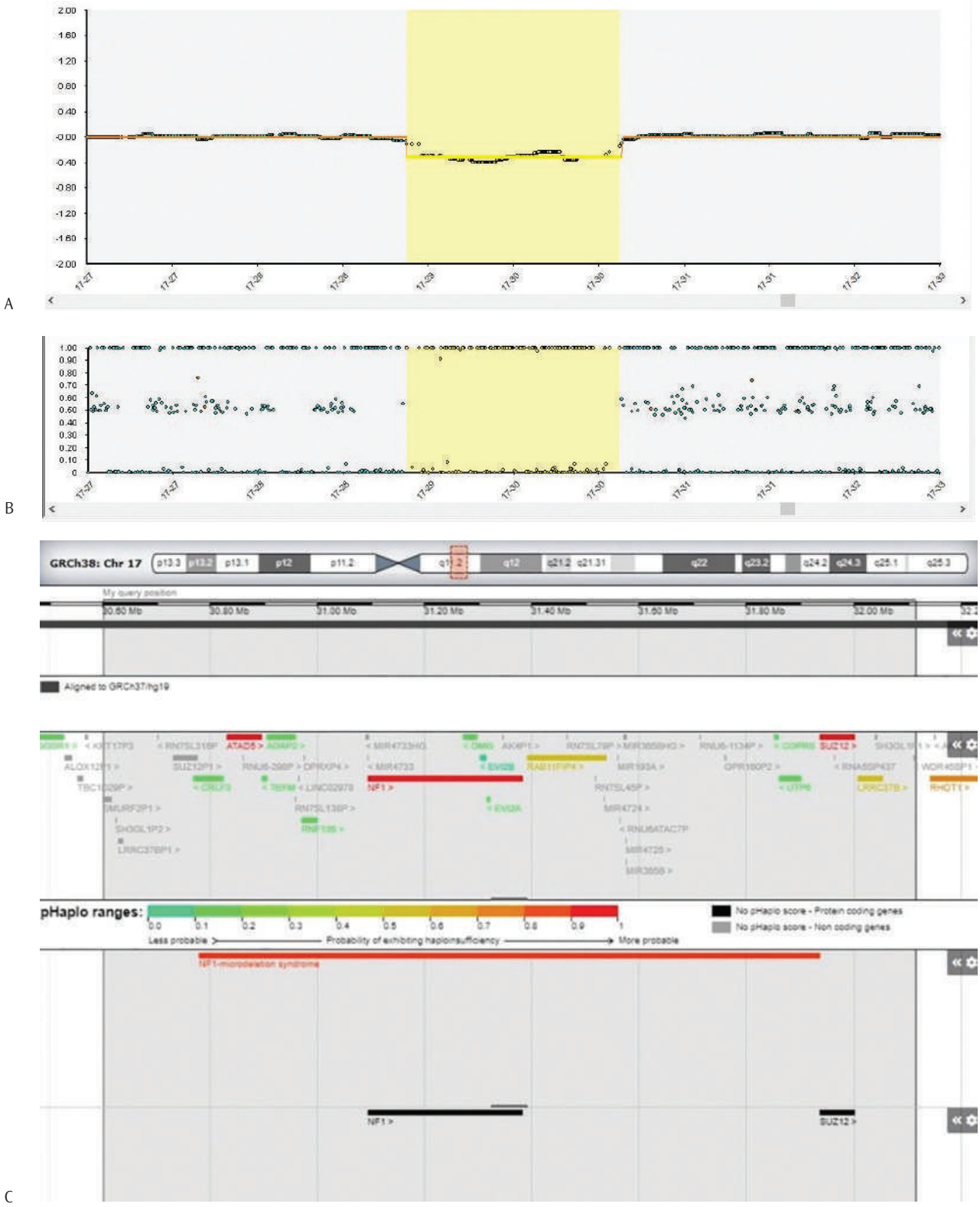

Figure 2. Microarray analysis of the patient. The log R chart (A) showed deletion of the region and the B-allele chart (B) revealed the absence of heterozygous allele peak in deleted region. The deletion (gray shaded area) included the "NF1-microdeletion syndrome region" and *NF1, SUZ12* genes which is shown by Decipher Genome Browser (C) (https://www.deciphergenomics.org/browser).

[OP-06]

Genetics of Charcot-Marie-Tooth Disease: Novel Variants and Rare Inheritances

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Abstract

Introduction: Charcot-Marie-Tooth disease (CMT) constitutes a heterogeneous group affecting motor and sensory neurons in the peripheral nervous system. The clinical features of CMT is variable due to genetic heterogeneity. To date, more than 80 causative genes have been reported. Autosomal dominant inheritance is common, while autosomal recessive inheritance and X-linked inheritance is rare.

Methods: We evaluated the genetic results of 5 cases who presented with a pre-diagnosis of CMT. NGS panel containing 55 genes was performed in four cases. WES was performed on a case with cognitive retardation in addition to CMT.

Results: Hemizygous pathogenic variants in *GJB1* gene was detected in two male cases. One variant [c.223C>T (R75W)] was a previously known variant, while the other [c.379A>C (I127L)] was novel variant. Both cases were evaluated as CMTX. A novel homozygous pathogenic c.2T>G(M1R) variant in the *NEFL* gene was detected in a male patient. The case was evaluated as the third case in the literature as autosomal recessive CMT1F. One of the two female patients had a known heterozygous pathogenic c.280C>T (R94W) variant in the *MFN2* gene, and the other had a novel heterozygous likely pathogenic c.455_456insG (D153Rfs*2) variant in the *GNB4* gene. These cases were evaluated as CMT2A2A and CMTDIF, respectively.

Conclusion: Genetic studies in CMT provide a wide range of benefits, from the identification of novel variants to the establishment of genotype-phenotype correlations. This study found three novel variants as the underlying causes of rare types of CMT with uncommon inheritance.

[OP-07]

Effectiveness of MediSom in Identifying Critical Fusions in Tumor Genome Profiling

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Abstract

Introduction: Accurate and efficient detection of gene fusions is critical for the proper management of cancer patient's diagnosis and treatment processes. This detection is often performed using bioinformatics software. The Istanbul Medipol University Center for Genetic Diseases Assessment Center aims to make significant contributions to this field with the MediSom Gene Fusion and SNP Detection pipeline. The capabilities of MediSom have been compared to internationally recognized bioinformatics programs in the detection of critical fusions in tumor genome profiles.

Methods: In this study, a comparative analysis was conducted on data from 17 cancer patients through the use of international bioinformatics software. Out of the 17 patient data, 3 were diagnosed with Sarcoma, and 14 were diagnosed with Lymphoma. Genetic data of patients were analyzed using the MediSom Gene Fusion and SNP Detection pipeline, and the results were compared with the results of 5 FISH studies.

Discussion: The study results demonstrated the successful detection of gene fusions in 15 patients using online software and MediSom. Among the fusion genes identified in Lymphoma patients were 5 cases of BCR-ABL1, 4 cases of ETV6-RUNX1, 2 cases of TCF3-BBX1, and 1 case of P2RY8-CRLF2. In Sarcoma patients, the detected fusions included EWSR1-FLI1, EWSR1-ERG, and PAX3-FOXO1 fusions. Additionally, FISH studies validated the results of MediSom in terms of fusion allele frequency, a critical measure reflecting the accuracy of detection algorithms.

Results: The MediSom Gene Fusion and SNP Detection pipeline have been observed as a valuable tool in the effective detection of gene fusions. When compared to international online software, a high level of concordance was observed. FISH studies further supported the reliability of MediSom. This study could provide valuable contributions to genetic diagnosis and treatment processes. In the future, a more detailed investigation into how MediSom performs in a broader range of patient groups and various genetic variations could shape advancements in this field.

[OP-08]

CGG Repeat Profile and Clinical Characteristics of Fragile X Full Mutant, Premutant and Gray Zone Patients

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Abstract

Introduction: Fragile-X Syndrome (FXS) is the most common cause of heritable intellectual disability (ID). It shows an X-linked inheritance pattern and more common in males than females. In addition to ID in full mutant (FM) patients, premutation carriers are at increased risk for diseases such as premature ovarian failure (POF) and Fragile-X Associated Tremor-Ataxia Syndrome (FXTAS). The aim of this study was to evaluate CGG in the *FMR1* gene genotype and phenotype profile of patients with repeat number 55 and above.

Methods: Ninety-two patients were included in the study. Genomic DNA was isolated from patient samples. CGG numbers with TP PCR test were determined. Patients were evaluated according to the ACMG guidelines for FM, premutant (PM) and gray zone.

Results: Forty-one patients were male and 51 were female. Thirty-four patients FM, 43 patients PM and 15 in the gray zone. Forty-four patients had size mosaicism. Of these patients, 16 had FM and 28 had PM. Expansion was generally found in CGG transmissions from mother to son. Contraction was found in father to daughter transmissions. Twenty-seven patients had attention deficit hyperactivity disorder and 22 patients had autism-spectrum disorder. The majority of these patients had FM. Eighteen patients had POF. Forty-four patients had a family history of early menopause and 93% of these patients were FM and PM. Two patients had findings compatible with FXTAS. In this disease caused by CGG repeat numbers with mitotic and meiotic instability, generational transmission and follow-up in terms of comorbidities are very important.

Keywords: Intellectual disability, Fragile X, premature menapouse

[OP-09]

Evaluation of Germline CHEK2 Variants and Challenges in Genetic Counseling

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Abstract

Inroduction: Germline variants in *CHEK2* gene have been associated with susceptibility to various cancers, especially breast and colon cancer. Although, it is known to be a gene with a moderate effective, it has been found a relationship between the type of mutations and cancer risk. For breast cancer, the odds ratio for truncating variants (eg. c.1100del, c.444+1G>A, ex8_9del) was found to be around 2.5, while for missense variants (eg. I157T, R117G, S428F, T476M) this ratio was around 1.5. Therefore, this situation should be taken into consideration when providing genetic counseling.

Methods: A custom-designed germline panel was applied to 580 patients with suspected familial cancer syndromes and results were evaluated with Qiagen QCI Interprete. All *CHEK2* gene variants detected in patients were collected and evaluated for mutation type and clinical significance based on Clinvar and Varsome data.

Discussion: A total of 18 different variants were detected in 53 patients in the cohort (9%). The E84E variant, which is a benign variant, was the most frequently observed variant with 37 patients 6.3%. Among the truncating mutations, c.444+1G>A (1 patient) and c.592+3A>T (3 patients) were detected, while various missense variants were detected in other cases. Of these, R180C, L183F, I157T, T476M were among the variants with pathogenic/likely pathogenic records in the Clinvar database. W93R, E478Q, D438Y, K141T, H371T, K373T, R519L, R137Q, S356L, E450K mutations were registered as of uncertain clinical significance in Clinvar. E84E and T532T are benign variants detected.

Conclusion: Various variants in the *CHEK2* gene are frequently encountered in daily cancer genetics practice. There is no clarity in the recommendations to be made in case of detection of some variants, especially missense variants, that have been detected throughout the gene and have previously been identified as disease-causing in databases such as Clinvar and in the literature, and this makes genetic counseling quite challenging.

Keywords: CHEK2, Truncating mutation, Familial cancer syndromes, Genetic counselling, Varaint of unknown significance

Introduction

The *CHEK2* gene was originally described in 1998 as an effector kinase in cell cycle regulation and ATM-CHK2-p53 DNA damage response pathway. Initial reports denoted the CHEK2 c.1100del variant could be causative in patiens with TP53-wt Li-Fraumani syndromes but following larger studies reported a high frequence (>1%) of this variant in healthy controls and lack of evidence for association to Li-Fraumeni syndrome (1).

Germline variants in *CHEK2* gene have been associated with susceptibility to various cancers, especially breast, colon and prostate cancers (2). Although it is considered that the *CHEK2* gene is a moderate risk gene for cancer predisposition, the risk is associated with the mutation type, family history, and modifying risk factors (3). Due to this uncertainity there is a big challenge for management of patients with CHEK2 germline pathogenic variants (GPV). Also with the emerging sequencing technologies, lots of variants with unknown significance were described among the genes (4).

Here, we report the distribution of all *CHEK2* gene variants in our cohort with tumor predisposition.

Materials and Methods

Patients referred for genetic analyses for familial cancer syndromes were enrolled in this study. DNA was extracted from peripheral blood with QIAamp® DNA Mini kit. Library preparation was achieved with a custom-design panel including all coding regions for *CHEK2* gene. High-troughput sequencing were done with Illumina Mi-Seq and Next-Seq platforms. Raw data were analyzed with QIAGEN CLC Workbench Premium and variants were interpreted with QIAGEN Digital Insights. All *CHEK2* gene variants detected in patients were collected and evaluated for mutation type and clinical significance based on Clinvar and Varsome data.

Results

A total of 580 patients with clinical suspicion of hereditary cancer syndromes including breast, ovarian and endometrium cancers were enrolled in this study. All the variants detected regardless of clinical significance were recruited. A total of 18 different variants were detected in 53 patients in the cohort (9%). A well-known benign variant, E84E, was the most frequently observed variant with 37 patients (6.3%). Also a benign variant T532T was detected in two patients (0.35%).

Two different truncating and 14 different missense variants were detected among the patients. Truncating mutations, c.444+1G>A was detected in a patient and c.592+3A>T was detected in 3 patients. Missense variants included R180C, L183F, I157T, T476M, W93R, E478Q, D438Y, K141T, H371T, K373T, R519L, R137Q, S356L, E450K. Among these missense variants, for R180C, L183F, I157T, T476M, there were both pathogenic/likely pathogenic and unknown significance citations in the Clinvar database. W93R, E478Q, D438Y, K141T, H371T, K373T, R519L, R137Q, S356L, E450K variants were registered as of uncertain clinical significance in Clinvar.

Discussion

Various variants in the *CHEK2* gene are frequently encountered in daily cancer genetics practice. There is no clarity in the recommendations to be made in case of detection of some variants, especially missense variants, that have been detected throughout the gene and have previously been identified as disease-causing in databases such as Clinvar and in the literature, and this makes genetic counseling challenging. Also there are many emerging new association efforts with various cancers (5). There have been a big challenge for genetic counseling in these patients. Clinical recommendation guidelines like "NCCN Genetic/Familial High-Risk Assessment: Breast, Ovarian, and Pancreatic Version: 2.2024" and "Genetic/Familial High-Risk Assessment: Colorectal Version: 1.2023" include parts concerning *CHEK2* gene. In these guidelines, it is described that primary breast cancer has 20-40% absolute risk and annual mammogram and breast magnetic resonance imaging (MRI) should be recommended. Also colorectal and prostate cancer risks are determined for CHEK2 variants. Also it is denoted that this risk is based only frameshift mutations and missense variants like I157T have lower risk ratios. So genetic counseling is unclear for missense variants (6).

A new practice resource have been published by "American Colage of Medical Genetics and Genomics". According to this paper, truncating variants were associated with the breast, colorectal and prostate cancers moderately. In the truncating mutations, mammogram and breast MRI from age 40 years for female breast cancer, self-examination for males breast cancer, measuring PSA levels for prostate cancers with family history have been recommended. For breast and colon cancer, missense variants are not clinically actionable in isolation and surveillance should be based on family history. For prostate cancer, it is not clear both the risk scores and management (7).

Conclusion

Lots of variants in *CHEK2* gene have been reported in the daily clinical genetic practice and there are lots of challenges about the genetic counseling. The latest published report is a candidate to remove the confusion on this issue. However, radical methods are not currently recommended for any of the CHEK2 variants.

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[OP-10]

Importance of SNP-Based Microarray Technology in Disorders of Sexual Development Work-up; Lessons Learned from a Male Case with 46,XX Karyotype

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Abstract

A 3-year-old male with 46,XX karyotype was referred to the medical genetics department. The post-partum manifestation was normal except micropenis, mid-penile hypospadias, and bilateral cryptorchidism findings. Thirty-seven weeks cesarean sectioned our patient's weight, height and body mass index percentile had been measured at -0.65 SDS, -1.01 SDS, and 0.2 SDS respectively. Ultrasonographic imaging confirms the testicular position without any atrophic or parenchymal abnormality with the right inguinal hernia. Normal Sertoli cell functions were revealed after serial anti-Mullerian hormone testing. A gonadal single dose stimulation test with 250 mcg/1.73 m^2 dosage of Ovidrel® revealed normal testosterone production as well. FISH-SRY revealed not any positive signals in 150 interphases and 5 metaphases first. SNP-CMA (>700K SNP probe) test was done utilizing GenomeStudio v2.0.5 software that confirms XX pattern in IGV. By using the chromosomal browser tab in the software, proximal abnormal patterns in the Y chromosome were detected. This pattern was different from other Y chromosomal patterns in the normal females with XX karyotype. Further SNP-Probe analysis shows the positiveness of the RPS4Y, ZFY, and SRY-associated SNP-Probes. By smooth filtering of the LOGR ratio, the very low mosaic pattern was revealed in the mentioned probes, which was later confirmed by the high metaphase FISH-SRY study. This case is showing the importance of gene(probe)-based approach analysis as well rather than the chromosomal or copy number variation-based approach only. On the other hand, it also clarifies the importance of the comparative (case-control) analysis approaches in the SNP microarrays technique.

[OP-11]

Investigation of the Effects of Conditioned Medium Derived from Amniotic Membrane on Proliferation in Cancer and Normal Cell Lines

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Abstract

Introduction: Human amniotic membrane (hAM), obtained after birth, is used in the treatment of various diseases due to its anti-angiogenic, anti-microbial and anti-proliferative properties. In recent years, researches have been focused on the anti-cancer properties of the amniotic membrane. However, reports on this subject are limited. In this study, we aimed to evaluate the anti-cancer effect of the conditioned medium (CM) derived from hAM on various cancer cell lines *in vitro*.

Methods: After cesarean section, placentas of healthy donor mothers were taken and amniotic membrane was obtained. hAM-CM was obtained by incubating amniotic membrane pieces with DMEM with 10% FBS and 1% PSA for 24 hours. HEK293, HUVEC and BV2 for normal cell groups; HEp2, PANC, MDA, A549 for cancer cell groups were cultured. To evaluate the cell viability of hAM-CM in normal and cancer cell lines, XTT test was performed by treating with 75% hAM-CM for 48 hours and the effects on the viability of cancer and normal cells compared to control cells were evaluated.

Results: According to the XTT results, after AM-CM treatment, the viability of normal cells decreased by an average of 13% ($p<0.05$), while there was also a decrease in the viability of cancer cells (HEp2, PANC, MDA, A549) by 4% (ns), 12% ($p<0.01$), 35% (ns), 18% ($p<0.001$), respectively.

Conclusion: Our results showed that the CM produced from hAM inhibited the proliferation of some cancer cells, while it would not be effective in others. hAM application for breast and lung cancer may be a new hope.

Keywords: Amniotic membrane, cancer, conditioned media, cytotoxicity, proliferation

Introduction

Cancer is one of the leading causes of death in the world (1). Despite conventional treatments such as surgery, chemotherapy and radiotherapy, cancer still has significant morbidity and mortality. Over the last decades many research studies have shifted their interests from conventional therapies to new therapies (2). Human amniotic membrane (AM) has recently attracted attention as an alternative treatment against cancer. The inner layer of the placenta is called amniotic membrane. AM, a biological barrier that supports the fetus, consists of amniotic epithelial cells, amniotic mesenchymal stem cells and fibroblasts (3). Due to its important biological properties, the AM has been used in medicine, especially in the field of ophthalmology and dermatology, for more than a century (4). In 1910, Davis used AM as a wound dressing for burn patients (5). Currently, AM derivatives are used in almost every field of medicine such as ophthalmology, plastic surgery, dermatology, cardiology, neurology, urology, diabetology, nephrology, pneumology, hepatology, transplantation, dental surgery, gynecology, orthopedic surgery and ear, nose and throat. There are more than 180 ongoing or completed clinical trials on this topic registered worldwide (6).

Different properties of AM, such as proapoptotic, anti-angiogenesis, cell cycle arrest, and immune regulatory, have made AM a suitable candidate for cancer-related research. In oncology, there is little research on AM, but it is anticipated that it may have more potential to be explored with further research (6). Seo et al. (7) first put forward a hypothesis on the potential of AM's anti-carcinogenic properties in 2008. Studies conducted so far have revealed the anti-cancer properties of AM through the secretion of different proteins and cytokines. Studies on the anti-tumoral effects of AM and cells in a limited number of cancer models indicate a potential anti-cancer effect of AM (8-10).

The anti-cancer mechanism of AM has not been clarified yet. Therefore, further studies are needed to elucidate the molecular mechanisms of the amniotic membrane, determine the factors involved in its anti-cancer effects, and translate amniotic cancer treatment into clinical practice. Because it is predicted that AM can be used as a new, safe and inexpensive substance with fewer side effects for cancer treatment in the future, due to its anti-inflammatory, anti-fibrotic, pro-apoptotic and anti-angiogenic effects. Thus, in this study we planned a pilot study aiming to investigate the anti-cancer effects of the human AM conditioned medium (hAM-CM) on normal and cancer cell lines. Our results may provide new pespectives about cancer treatment.

Materials and Methods

Placental Tissue Collection and Preparation of Amniotic Membran Condition Medium:

After obtaining informed written consent from all subjects according to the guidelines set by the Ethical Committee of Selçuk University of Medical Sciences, placentas were collected from Selçuk University Faculty of Medicine, Department of Obstetrics and Gynecology. Placentas (40 weeks) from healthy donor mothers (n=6) after caesarean section from normal term pregnancies were harvested, suspended in cold PBS and transported to the laboratory. The AM was manually stripped and cut from the chorionic membrane and placed in sterile physiological solution. Bloody or torn pieces of the AM was washed two or three times to completely remove them and then cut into 4 equal pieces. Each piece will be sterilised in a laminar flow by continuous washes in PBS containing 100 U/mL penicillin and 100 µg/mL streptomycin. Amnion pieces were placed in 10 cm plates and incubated with DMEM with 0% FBS for 24 hours. After removing the AM fragments, the remaining medium is centrifuged at 1,200 rpm for 5 min to remove cellular debris, after which the cell pellet was discarded; the remaining medium is termed human conditioned medium (hAM-CM) and was pooled from 10 different placenta donors and stored at -20 °C until used in experiments.

Cells and Cell Culture

HEK293 (human embryonic kidney cells), HUVEC (human umbilical vein endothelial cells), BV2 (mouse microglial cells), HEp2 (human larynx cancer cell), PANC (human pancreatic cancer cells), MDA (human breast cancer cells), A549 (human lung cancer cells) were purchased from the American Type Culture Collection (ATCC). Both of the cells were maintained in Dulbecco's Modified Eagle's Medium (DMEM) (Gibco, USA) supplemented with 10% fetal bovine serum (FBS) (Gibco, USA) and 1% penicillin/streptomycin (Gibco, USA) in a humidified 5% CO_2 incubator at 37 °C.

Cell Proliferation Assay

Cells were seeded into 96-well plates ($1x10^4$ cells per well). After overnight attachment, the medium was replaced with hAM-CM diluted in a ratio of 3:4 (75%) with a complete medium for 48 h. An untreated group of cancer and normal cells that incubated with a complete medium was considered as a control group. Cell viability was assessed at 48 h. The XTT kit (BI, USA) was used according to the manufacturer's instructions. Results were obtained by detecting absorbance at a wavelength of 540 nm for formazan and 650 nm for background with the microplate reader (Biotek-Epoch, USA). Three replicate wells were used for each analysis. The percentage of cell viability (%) compared to the untreated cells was determined at each concentration using the formula provided below:

$$\text{Cell viability (\%)} = \frac{\text{OD of sample}540 - \text{OD of sample}650}{\text{OD of control } 540 - \text{OD of control}650} X100$$

Morphology Analysis

To determine the effect of co-culture on cell morphology, treated/untreated normal and cancer cells were examined under an inverted light microscope (Nikon) after 48 h before XTT assay.

Statistical Analysis

Data were presented as the mean ± standard deviation of three independent assays, and data were analyzed using the GraphPad Prism software version 6.00 for Windows (GraphPad Software, USA). The student's t-test was used to perform statistical analysis. Differences were considered significant at $p<0.05$.

Results

Effects of hAM-CM on Cell Morphology

In AM-CM-treated cell groups, only the morphology of microglia cells (BV2) differed compared to control cells. The connection between cells was broken after treatment. On the other hand, no significant changes were observed in the morphology of other cells (Figure 1A). There is no significant morphological differentiation in the cancer group compared to the control groups, except for breast cancer cells. However, breast cancer cells have severe morphological differentiation as if under stress (Figure 2A).

Effects of hAM-CM on Cell Proliferation

According to the XTT cell viability assay results, there was a decrease in cell viability in both healthy and cancer cells when compared to untreated control cells. In normal cells (HEK293, HUVEC, BV2), there was a decrease of 13% (ns), 14% ($p<0.01$), and 11% ($p<0.01$), respectively (Figure 1B). In cancer cells (HEp2, PANC, MDA, A549), there was a decrease of 4% (ns), 12% ($p<0.01$), 35% (ns), and 18% ($p<0.001$), respectively (Figure 2B).

Discussion

Using hAM has shown encouraging results in the treatment of various diseases; but, their impact on cancers is contradictory and insufficient. Thus, we investigated the anti-cancer properties of the conditioned medium derived from the human AM on cancer cells to determine whether AM produce paracrine mediators that affect cancer cell culture growth.

To be sure whether the effect of AM-CM on cancer cells is specific or not, we also evaluated its effect on normal cells. For this reason, we studied laryngeal, pancreatic, breast and lung cancer cells, as well as epithelial (HEK), endothelial (HUVEC) and microglia (BV2) cells. As a result, the viability rate of normal cells was 87% on average, which was lower than the viability rates of MDA (65%) and A549 (82%) cells compared to cancer cells. In the study of Rolfo et al. (11), AM did not inhibit the proliferation of normal prostate cells. Based on these results, we can conclude that the anti-cancer activity of AM is cell dependent. Therefore, by treating AM in different cell lines, the cells in which AM is most effective can be investigated. Mamede et al. (3) treated prostate, breast, colon, lung, pancreatic, biliary, endometrial, bladder, esophagus, liver, bone and skin cancer cells with proteins derived from AM (AMPE). Accordingly, they proved that some cancer cell lines are much more sensitive to AMPE. This result is in accordance with our data that the inhibition of metabolic activity by AMPE may differ even between different cell lines of the same cancer type, depending on the specific genetic profile of each cell line (3). Therefore, it is important to study the individual response of each cell line.

Jafari et al. (10), in their study, demonstrated inhibition of cell proliferation in breast cancer cell lines treated with AM-CM. According to our data, among the four cancers we studied, the inhibitory effect of AM-CM was highest in breast cancer.

Rolfo et al. (11) studied with CM which was produced by incubating mesencimal cells derived from mesencimal cells (hAMSC) with DMEM medium for 3 days and 1 week. Their data indiacted that hAMSC microenvironment inhibited prostate cancer cells growth. In our work, during the production of AM-CM, amniotic fragments were incubated in the medium for 24 hours. If this period is increased to 48 or 72 hours or more hours, it will be clear whether the activity of AM-CM on cancer cells will increase or not.

In the literature, different forms of AM such as whole membrane, fresh, dried or freeze-stored, and AM-derived epithelial cells and mesenchymal cells have also been used (12-14). In this study, we used conditioned media obtained from fresh whole membrane. In future studies, the effect of these forms of AM on cancer cells can also be evaluated.

AM has been reported to have many functions such as anti-angiogenic, anti-fibroblastic, proapoptotic, anti-microbial, immunomodulatory activities as well as enhancing cell migration and growth (15-18). In our study, we investigated the effect of AM on cell growth. We aim to investigate other effects such as migration, metastasis, apoptosis in future studies.

Conclusion

Our data report that conditioned media delivered from human AM probably contains soluble factors responsible for triggering an anti-tumor respons. But its effect is cell dependent.

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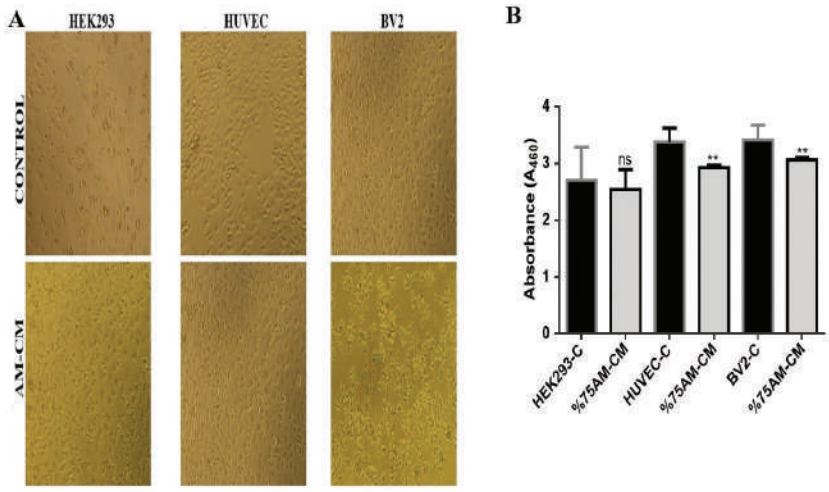

Figure 1. The effect of hAM-CM treatment on cell morphology and proliferation in normal cells. (A) The morphology in control, and treated groups of HEK293, HUVEC and BV2 cells. (B) hAM-CM decreases cell proliferation in HUVEC and BV2 cells (**p<0.01).

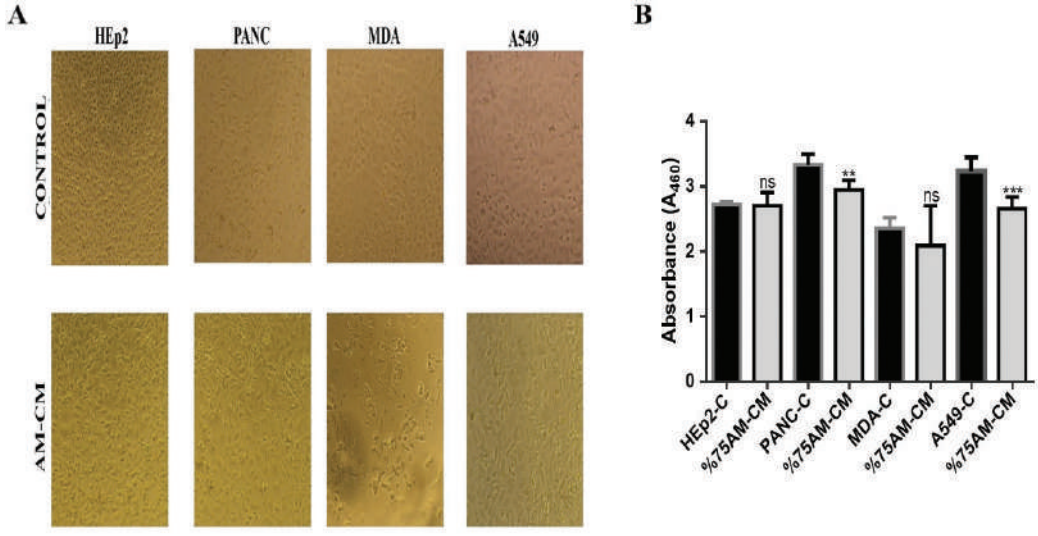

Figure 2. The effect of hAM-CM treatment on cell morphology and proliferation in cancer cells. (A) The morphology in control, and treated groups of HEp2, PANC, MDA and A549 cells. (B) hAM-CM decreases cell proliferation in PANC, MDA and A549 cells (**p<0.01; ns and ***p<0.001).

[OP-12]

Molecular Diagnosis in Spinocerebellar Ataxias

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Abstract

Introduction: Spinocerebellar ataxias (SCA) constitute a genetically and phenotypically heterogeneous group of progressive neurodegenerative diseases inherited in an autosomal dominant. Mutations causing the disease often result from the expansion of tandem repeats within the disease gene, leading to abnormal elongation of polyglutamine. Among the disease mechanisms of SCAs are toxic RNA gain-of-function, mitochondrial dysfunction, channelopathies, autophagy, and transcriptional dysregulation. In addition, among the autosomal recessively inherited cerebellar ataxias, there are SCA affecting extraneurologic systems with onset of symptoms especially in early childhood.

Methods: A total of 160 patients who presented to our clinic with a preliminary diagnosis of SCA between 2019 and 2023 were included in the study. A panel including SCA 1, 2, 3, 6, 7, and 8 were studied through short tandem repeat analysis. Whole exome sequencing (WES) was performed on two patients with normal results.

Results: Among the 160 patients evaluated with SCA symptoms (female/male: 77/83, average age: 36.4), an increase in repeat numbers explaining clinical findings was detected in 31. Of these patients, 25 were diagnosed with SCA type 1, 5 with type 2, and 1 with type 8. Two patients who were found to have normal results and underwent WES were diagnosed with SCA type 8 (MIM: 610743) and Spastic Ataxia, Charlevoix-Saguenay (MIM: 270550).

Discussion: In this study, the most common types were diagnosed in 19.3% of patients with a prediagnosis of SCA. While types 3, 2, 6, and 1 are most commonly reported in the literature, our study observed types 1 and 2 as the most frequent. Evaluating family history and additional systemic involvement in patients with symptoms starting early is crucial for determining tests for diagnosing other SCA types. Currently, there is no cure for SCAs, but knowing the subtype can assist in managing symptoms, determining treatment strategies to improve quality of life, and accessing preimplantation/prenatal diagnosis options to prevent the disease.

Keywords: Spinocerebellar ataxia, short tandem repeat analysis, whole exome sequencing

[OP-13]

Change in Lipid Metabolism Gene Expressions during TGF-β-Induced Epithelial-Mesenchymal Transition Process

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Abstract

Introduction: Epithelial-mesenchymal transition (EMT) is a biological process where cells lose their epithelial characteristics and acquire mesenchymal features. EMT plays a critical role in normal development and cancer metastasis. During EMT, lipid metabolism meets cellular energy demands, contributes to structural changes, and supports metastasis. Transforming growth factor-beta (TGF-β) signaling pathways initiate EMT and regulate the transcription of genes influencing lipid metabolism. This study aims to examine changes in lipid metabolism gene expressions during TGF-β-induced EMT and identify candidate genes that may play a role in this process.

Methods: In this study, Py2T breast cancer cells derived from MMTV-Py2T transgenic mice were cultured to induce a mesenchymal phenotype with TGF-β. RNA isolation was performed from Py2T and Py2T-TGFβ cells, followed by a targeted RNA sequence analysis. The study focused on 180 genes involving adipogenesis, lipogenesis, lipid storage, β-oxidation, MEK/ERK pathways, and also examined interleukin gene expressions. Gene pathway analysis was conducted using various bioinformatics tools.

Discussion: These results demonstrate the complexity of lipid metabolism gene expressions during TGF-β-induced EMT and the significance of cellular phenotypes in this process. Additionally, genes such as Trib3, Ceacam1, Bmp2, and Itgb3 may be associated with EMT and could impact cancer prognosis. Future studies could further support these findings by examining the functions of these genes in more detail.

Results: According to the bioinformatics analysis results, significant changes in expression levels were detected in a total of 29 genes, including important genes like Fasn, Cebpa, and Egr1. In comparison to cancer epithelial cells, cancer mesenchymal cells showed a decrease in Trib3 (-2.7-fold) and Ceacam1 (-3.7-fold) gene expression levels and an increase in Bmp2 (5-fold) and Itgb3 (4.4-fold) gene expression levels ($p<0.05$).

Keywords: Breast cancer, EMT, lipid metabolism, TGF-β

[OP-14]

Evaluation of Genetic Heterogeneity in Joubert Syndrome Case Series

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Abstract

Introduction: Joubert syndrome is a primary ciliopathy disorder which is inherited as autosomal recessive and characterized with cerebellar and brainstem malformations. In addition to being a clinically heterogeneous disease with multisystemic involvement, it is also genetically heterogeneous as more than 39 genes can be involved in the etiology. Molar teeth sign due to cerebellar vermis hypoplasia on brain magnetic resonance imaging is pathognomonic; and hypotonia, developmental delay, intellectual disability are among the diagnostic criteria. In this study, the results of patients who were referred to our center with a preliminary diagnosis of Joubert syndrome and underwent molecular genetic examinations were presented and evaluated in terms of genotype-phenotype correlation. It was aimed to raise awareness about Joubert syndrome, which is relatively common among rare diseases.

Methods: Among the patients included in the study, in 7 of them targeted genes (*CPLANE1, NPHP1, TMEM237, KIAA0586, TMEM67, CC2D2A, HYDIN, KIF7, AHI1, ARMC9*) were sequenced via Next Generation Sequencing Method and Whole Exome Sequencing was performed in the other 16 patients.

Results: As a result of the studies, homozygous mutations were detected in 7 patients, compound heterozygous mutations in 7 patients, and heterozygous mutations in 9 patients. A total of 21 different variants were detected in 10 different genes. As a result of the evaluation according to ACMG criteria, 15 variants were classified as pathogenic, 5 variants of unknown clinical significance, and the other 1 variant was not previously identified and was classified as possible pathogenic as a result of *in silico* analyses. The c.1466+2T>A variant in the *CC2D2A* gene is the novel mutation to be introduced into the literature. The gene with the most frequent mutation is revealed as *KIAA0586* gene.

Discussion: By revealing the etiopathogenesis of rare diseases, the possibility of early diagnosis, prenatal and pre-implantation genetic diagnosis increases, and preventive medicine can come to the fore. With the increase in similar studies, it will be possible to create national databases and develop new algorithms for diagnosis and treatment.

Keywords: Rare disease, Joubert syndrome, mutation, whole exome sequencing

[OP-15]

A Case of Angelman Syndrome with c.2570_2571delAA Variant in *UBE3A* Gene, and Unusual Findings

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Introduction

Angelman syndrome is a neurodevelopmental disorder characterized by severe developmental delay or intellectual disability, profound speech impairment, movement or balance disorder, typical abnormal behaviors with an apparent happy demeanor that includes frequent laughing, smiling, and excitability. Microcephaly and seizures are also common. Four known genetic mechanisms may cause Angelman syndrome; deletion of the 15q11.2-q13 critical region (60-75%), paternal uniparental disomy (2-5%), imprinting defect (2-5%) and mutation in the *UBE3A* gene (10%). In a group representing 5-26% of patients, the genetic defect remains unidentified. Here, we present a case with Angelman syndrome with unusual clinical findings, and likely pathogenic variant in *UBE3A* gene.

Case Report

Seven-years-old male patient was evaluated for developmental delay. He could not speak, his 13 teeth were extracted because of caries, he slept all day, and he could stay awake with treatment, he had sensory integration defect. On examination, he had microcephaly, deep set eyes, macrostomi, scoliosis, tremor on right hand, and wide-based gait. His karyotype, microarray, 15q11.2-q13 methylation analysis were normal. Thereafter we have performed the sanger sequencing analysis of *UBE3A* gene. We found a heterozygous likely pathogenic frameshift c.2570_2571delAA(K857Tfs*2) variant (NM_000462.5). The parents had normal sequencing for the same position.

Discussion

Despite the sleep problem was contrary to the literature, electroencephalogram was normal, hypopigmentation and happy puppet appearance and typical position were absent, the diagnosis was made with some remarkable clinical features initially. This rare case is presented to emphasize the importance of proceeding with clinical findings, and to contribute to the phenotype-genotype correlation.

[OP-16]

Rare Syndrome Caused by a Novel Splice Variant in *UBE2A*: A Case Report

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Introduction

X-linked intellectual disabilities are responsible for approximately 10-12% of intellectual disability cases in males. Nascimento type X-linked syndromic cognitive developmental disorder (MRXSN) is one of these conditions. Mutations in the *UBE2A* gene and large Xq24 deletions encompassing *UBE2A* are responsible for this condition. The *UBE2A* gene encodes the E2 enzyme (ubiquitin-conjugating enzyme) involved in attaching ubiquitin to proteins. In MRXSN, in addition to intellectual disability, individuals may exhibit seizures, speech difficulties, dysmorphic facial features, skin abnormalities, and urogenital system anomalies.

Case Report

A 6-year-old male patient, who is being followed up for diagnoses of epilepsy and global developmental delay at the Neurology Department, is being consulted. He has been experiencing seizures since the age of 11 months, occurring approximately once a year. These seizures are characterized by the patient staring at a single point and experiencing atonia in the body. The patient was born to healthy, non-consanguineous parents after an uncomplicated 38-week pregnancy. Sitting without support started at age 1.5 and walking in the form of steps was achieved at the age of 4. The child has never developed speech and only produces non-sensical sounds. He understands simple commands. In physical examination; obese appearance, broad face, synophrys, deep-set almond-shaped eyes, wide mouth, short neck, broad and flat nasal bridge, widely spaced nipples, micropenis, onychodystrophy in toenails, and hirsutism on the back and arms were observed. Additionally, bilateral frontal paroxysmal findings were detected in electroencephalography (EEG). Due to the syndromic appearance, microarray analysis was performed on the patient, which yielded normal results. In the whole exome analysis, the hemizygous c.241+5G>A variant was detected in the *UBE2A* gene and was interpreted as likely pathogenic. This variant was also identified as heterozygous in his mother. Segregation analysis of other family members is ongoing.

Discussion

MRXSN, initially described in 2006 by Nascimento and colleagues, was identified in three individuals diagnosed with cognitive impairment, where a non-sense mutation in *UBE2A* was detected. In addition to that, in cases reported so far, large and small deletions, various missense mutations, an insertion, and two splicing variants have been identified. These splicing variants were identified in a 5-year-old Chinese patient and two Italian brothers aged 26 and 36. In the case of the 5-year-old patient, cognitive impairment, speech disorder, and white matter anomalies were observed. However, due to the absence of dysmorphic facial features, seizures, skin anomalies or, urogenital system abnormalities, this case was considered mild. In contrast, dysmorphic facial features are evident in Italian siblings. In addition to the neurodevelopmental features of the syndrome, skin and urogenital system anomalies, serious behavioral disorders have been observed.

Conclusion

The distinctive dysmorphic facial features, skin, nail, and urogenital anomalies, intellectual disability and neurodevelopmental delay observed in MRXSN patients suggest that this syndrome presents clinically recognizable characteristics. In cases of undiagnosed intellectual disability in male patients, differential diagnosis may consider *UBE2A*-related MRXSN based on a detailed medical history, physical examination, and family history. With this rare case, which has been identified in 20 families in the literature so far, it is aimed to better understand the characteristics of the syndrome and demonstrate the impact of the observed novel variant on the phenotype.

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[OP-17]

Neuromuscular Diseases, Genetic Etiology and Next Generation Sequence Analysis

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Abstract

Introduction: Neuromuscular diseases (NMD) are characterized by the deterioration of muscles nerves building blocks of the movement system. Non-congenital acquired NMD may occur due to environmental factors, infections, side effects of various pharmacological treatments. On the other hand, gene variants that affect muscle-nerve structure function have a major role, which has a genetic or hereditary etiology.

Methods: This study aims to investigate NMD using the next generation sequence (NGS) analysis method from patients who applied to the Department of Medical Genetics with the prediagnosis of neuromuscular involvement between the years 2020-2023. In our study, more than 200 NMD-related gene NGS methods were analyzed in twenty-four pre-diagnosed patients with NMD.

Discussion: NMD is classified as anterior horn, peripheral nerve, neuromuscular junction and muscle diseases. In the classification, spinal muscular atrophy, amyotrophic lateral sclerosis, Guillain Barre syndrome, Charcot-Marie-Tooth, congenital myasthenic syndromes, muscular dystrophies are among the prominent ones. DNA sequence analysis is extremely important of NMD diagnosis algorithm of patients. Therefore, it becomes possible to perform NMH typing in such a wide spectrum and to target gene therapy with the detection of the variant, as in Duchenne muscular dystrophy. The novel variants will contribute to the development of differential diagnosis and treatment strategies, combination of clinic and genetic diagnosis will reveal the definitive diagnosis.

Conclusion: NMH-related VUS, pathogenic variant was detected in patients in the study, a clinically compatible genetic diagnosis of the patients was determined. For variant zygosity and novel detected variants, segregation analysis was performed, genetic counseling was given for risky individuals and carriers in the family.

[OP-18]

Genetic Epilepsy and Molecular Diagnosis

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Abstract

Introduction: Epilepsy is a neurodegenerative disease with phenotypic and genotypic heterogeneity. While a single gene mutation may be responsible for etiopathology, epilepsy types with familial inheritance or predisposition may also show complex inheritance together with environmental factors. In parallel with the development of molecular genetic techniques, epilepsy-related gene variants can be identified with next generation sequence (NGS), the inheritance of the disease can be revealed and genetic counseling can be given with risk assessment.

Methods: This study aims to investigate the genetics of epilepsy using the NGS analysis method from patients who applied to the Department of Medical Genetics with a prediagnosis of epilepsy between the years 2020-2023. Early infantile epileptic/encephalopathy panels were studied through Minieq sequencing system. The pathogenicity classification of the detected variants over a hundred thirty patients was determined together with the clinical diagnosis. The segregation analyses were performed according to the zygosity, in line with their pedigrees.

Discussion: Rapid advances in NGS have evolved to exciting results in epilepsy genetics. The combination of epilepsy-related gene panels and clinical tests has become more common, leading to more effective diagnoses, especially in early-onset epilepsy. This diagnostic algorithm sheds light on the pathophysiology of both early-onset benign and epileptic syndromes and devastating developmental and epileptic encephalopathies.

Conclusion: Among the patients included in the study, variant classification was made according to pathogenicity scoring according to NGS results, and segregation was determined in patients with novel or homozygous variants. In line with the results, molecular genetic diagnosis and genetic epilepsy determination of the patients were revealed.

[OP-19]

5-Aza-2'-Deoxycytidine May Suppress Prostate Cancer Cell Proliferation by Altering The Expression of miRNAs

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Abstract

Introduction: Prostate cancer (PCa) is one of the most common and lethal malignant tumors. 5-Aza-2'-deoxycytidine (Aza) is a methylation inhibitor and an essential drug used in the treatment of different malignancies by modifying the expression of several genes. In our study, the effects of Aza on cellular processes and expression of miR-15a-5p, miR-16-5p, and miR-7-5p in PCa cells were investigated.

Methods: DU145 and PC3 PCa cells were treated with 10 µM Aza (Aza+), and control cells were treated with 10 µM DMSO (Aza-). The effect of Aza on PCa cell proliferation was determined using the WST-8 method, and the effect on apoptosis was determined using the Caspase-3 assay kit. examined. The migration potential of cells was evaluated using a scratch assay. cDNA was then synthesized from the total RNA samples obtained from Aza(+) and Aza(−) cells. MiR-15a-5p, miR-16-5p, and miR-7-5p were quantified by qRT-PCR using the TaqMan primer-probe assay.

Results: In DU145/Aza(+) and PC3/Aza(+) cell lines, proliferation, and migration were significantly decreased and apoptosis was increased in Aza(+) cells compared to Aza(−) cells. In addition, decreased expression of miR-15a-5p and miR-16-5p was detected in DU145/Aza(+) and PC3/Aza(+) cells compared to the control group, while increased miR-7-5p expression was detected.

Discussion: Azacitidine suppressed the proliferation and migration of PCa cells and induced apoptosis. Azacitidine may have affected these cellular processes by directly miRNA hypomethylation or indirectly by changing the expression of miR-15a-5p, miR-16-5p, and miR-7-5p through different target genes.

Keywords: Prostate cancer, DU145, PC3, 5-Aza-2'-deoxycytidine, miRNA

Introduction

Prostate cancer (PCa) is the second most common cancer among men. It has been reported that the combined application of 5-Aza with various chemotherapeutics (such as docetaxel) in the treatment of PCa has a positive contribution to the increase in treatment sensitivity, especially in chemotherapy-resistant patients (1). MicroRNAs (miRNAs) are small molecules. Although non-coding, they are crucial molecules because they act as regulators of the coding genes. miRNAs targeting oncogenes are called tumor suppressor miRNAs (Ts-miRs), while those targeting tumor suppressor genes are called Onco-miRs (2). 5-Azacitidine is an effective but toxic methylation inhibitor. 5-aza-2'-deoxycytidine (also known as 5-Aza or decitabine) was synthesized by reducing the toxicity of this molecule. This agent is used to demethylate methylated genes and non-coding RNAs that regulate gene expression for therapeutic purposes in cancer (3). It has been suggested that miR-15a and miR-16-1, which are detected at reduced levels in the tumor tissue and serum of patients with PCA, may be non-invasive biomarkers for the diagnosis and prognosis of PCa (4-6). In addition, miR-7 expression has been reported to decrease in drug-resistant PCa tissues (7). In our study, the effects of Aza on cellular processes and the expression of miR-15a-5p, miR-16-5p, and miR-7-5p in PCa cells were investigated.

Materials and Methods

Culture of DU145 and PC3 cells and treatment with 5-Aza-2'-deoxycytidine: DU145 and PC3 cells were cultured in RPMI1640 medium containing 10% FBS and 1% antibiotics at 37°C and 5% CO2. The 50% inhibitory concentration (IC50) of 5-Aza in DU145 and PC3 cells was determined as 10µM. DU145 and PC3 PCa cells were treated with 10µM Aza (Aza+), and control cells were treated with 10µM DMSO (Aza-). The effect of 5-Aza-2'-deoxycytidine on PCa cell proliferation, apoptosis, and migration: For the proliferation assay, cells were seeded in triplicate in a 96-well plate. On the other hand, cells were seeded on a 6-well plate for apoptosis and migration assays. The effect of Aza on PCa cell proliferation was investigated using the WST-8 method (Ecotech Biotech), and the effect on apoptosis was determined using the Caspase 3 Human Instant ELISA Kit (Invitrogen) according to the manufacturer's protocol. The effect of Aza on PCa cell migration was examined using the scratch assay. PCa cells were seeded in 6-well plates (4×105 cells/well). Scratches were performed using a 100 µL pipette tip when the cells reached 100% confluency. After 48 h, the scratch closure potentials of the Aza (+) and Aza(−) cells were compared under an inverted microscope. cDNA synthesis and expression analysis: cDNA was synthesized from 30ng/µl total RNA samples obtained from Aza(+) and Aza(−) cells. MiR-15a-5p, miR-16-5p, and miR-7-5p were quantified by qRT-PCR using a TaqMan primer-probe assay (Thermo Fisher).

Statistical Analysis

The student's t-test and the 2-ΔΔCt method were used for statistical data analysis. P value less than 0.05 was considered significant. GraphPad Prism 9.5.1 program was used to create the figures, and values are indicated as $p \leq 0.05$ (*), $p \leq 0.01$ (**), and $p \leq 0.001$ (***).

Results

When the effect of 5-Aza on cellular processes in DU145/Aza(+) and PC3/Aza(+) cells was observed; it was detected that proliferation (Figure 1) and migration (Figure 2) were significantly decreased and apoptosis (Figure 3) increased in Aza(+) cells compared to Aza(−) cells.

In addition, decreased miR-15a-5p (p=0.013; p=0.003, respectively) and miR-16-5p (p=0.003; p=0.011, respectively) expressions were detected in DU145/Aza(+) and PC3/Aza(+) cells compared to the control group, while increased miR-7-5p (p=0.002; p=0.0012, respectively) expression was detected (Figure 4).

Discussion

It has been understood that 5-Aza-2'-deoxycytidine suppresses the proliferation and migration of PCa cells and induces apoptosis. This may have affected these cellular processes by direct miRNA hypomethylation or indirectly by altering the expression of miR-15a-5p, miR-16-5p, and miR-7-5p through different target genes.

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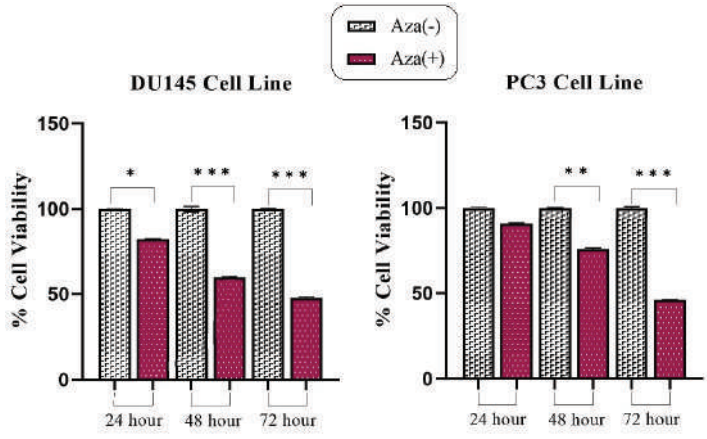

Figure 1. The effect of 5-Aza-2'-Deoxycytidine on PCa cell proliferation

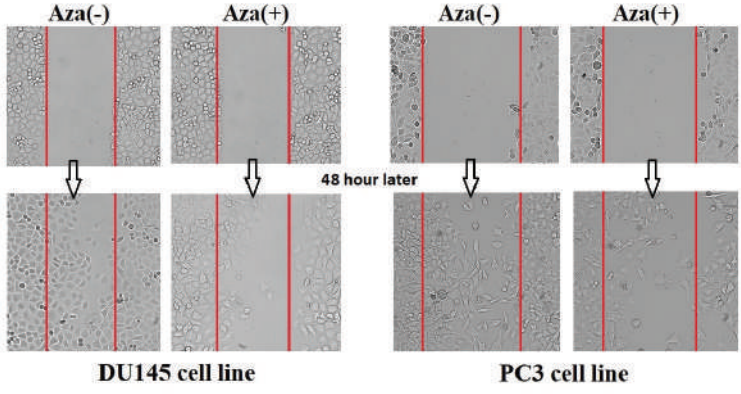

Figure 2. Cell migration (scratch) assay of PCa cells treated with 5-Aza

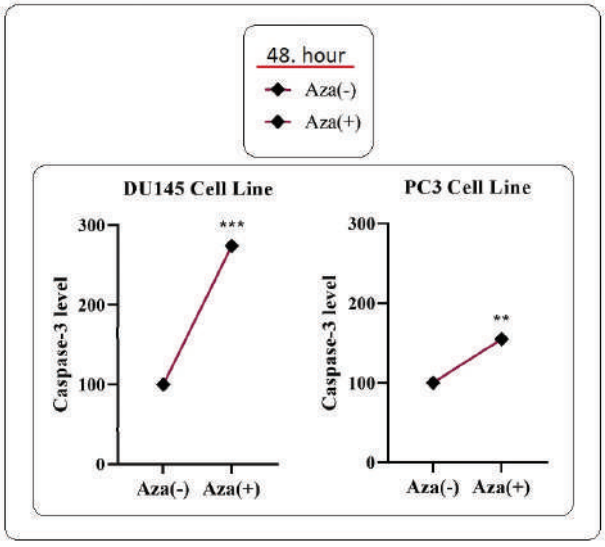

Figure 3. Apoptosis assay result for DU145/Aza(+) (p=0.00017) and PC3/Aza(+) (p=0.002) cell lines

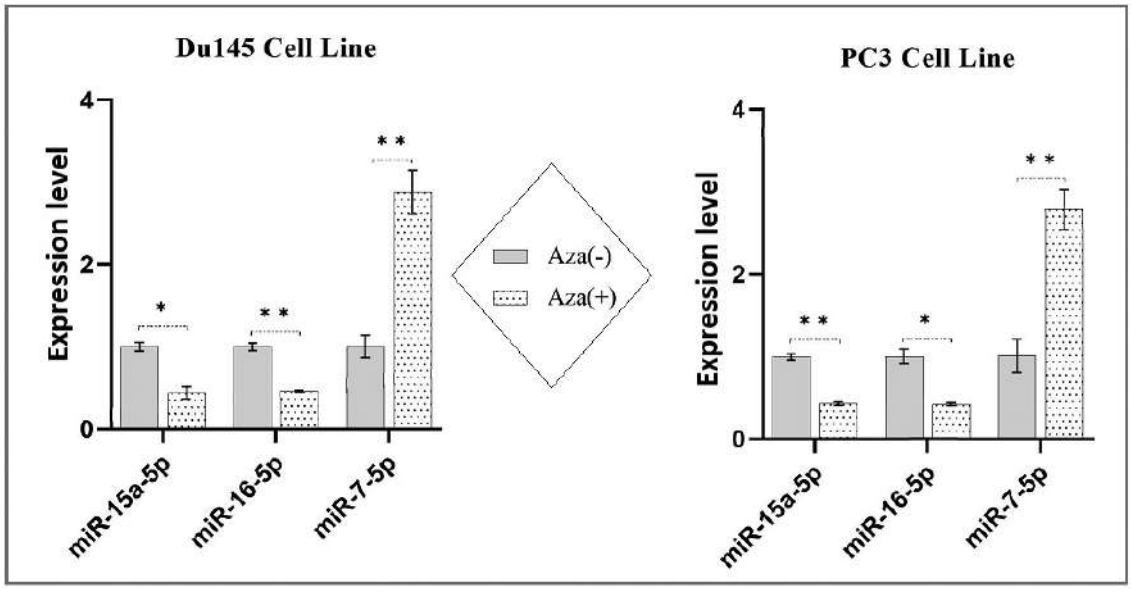

Figure 4. Relative expression level of miR-15a-5p, miR-16-5p and miR-7-5p in Aza(+) and Aza(-) cell

[OP-20]

Investigation of *NF-κB* Gene Polymorphisms in Individuals Infected with COVID-19 Virus

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Abstract

Introduction: Coronavirus disease-2019 (COVID-19) is a disease caused by the severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) coronavirus that emerged in Wuhan, China. SARS-CoV-2 is an enveloped, positive-sense, single-stranded RNA virus belonging to the betacoronavirus genus. NF-κB is a family of protein transcription factors that can orchestrate many inflammatory processes. NF-κB genes have been associated with the development of many viral diseases and cancer. Therefore, this study aimed to investigate COVID-19 disease and NF-κB1-94 Ins/Del ATTG, NF-κB1A 3'UTR A/G, NF-κB1A -826 C/T polymorphisms.

Methods: The patient group was composed of 150 individuals who were definitively diagnosed with COVID-19 disease by Sivas Cumhuriyet University Faculty of Medicine, Department of Infectious Diseases. Approval was received by Sivas Cumhuriyet University Clinical Research Ethics Committee (decision no: 2021-02/07). The control group consisted of 150 healthy individuals whose blood was taken during the period when there was no COVID-19 epidemic (Ethics Committee decision no: 2009-02/5). Total DNA was isolated from the blood samples taken, and then PCR-RFLP study was performed to determine polymorphisms. The data were uploaded to the SPSS (Ver: 23.0) program and the chi-square test was used for evaluation.

Results: A statistically significant difference was found in the D allele distribution for the NF-κB1 -94 ins/del polymorphism. Additionally, a statistically significant result was found in the WD and DD genotype distribution. For NF-κB1A 3'UTR A/G polymorphism, there was a statistically significant difference in the distribution of the G allele of patients with COVID-19. Additionally, CT and TT genotype distributions were found to be statistically significant. The T allele distribution for the NFk-B1A -826 C/T polymorphism has a statistically significant difference, and the CT and TT genotype distributions were also found to be statistically significant.

Conclusion: There is a significant relationship between COVID-19 disease and NF-κB genes, but further studies are needed for this gene.

Keywords: NF-κB, COVID-19, polymorphism

Acknowledgments: This study was supported by CUBAP project no: F-2021-635.

[OP-21]

Mosaic Paternal Uniparental Disomy of chromosome 11 in a Patient with Beckwith-Wiedemann Syndrome

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Abstract

Beckwith-Wiedemann syndrome (BWS) is an imprinting disorder that leads to overgrowth and increased risk of embriyonic tumors. It is associated with genetic and epigenetic changes on the chromosome 11p15 region, which includes imprinted genes that regulate fetal and postnatal growth. BWS is considered a spectrum (BWSp) ranging from classic BWS to isolated lateralized overgrowth. A 5-year-old girl patient was referred to us from the neurology outpatient clinic due to speech retardation. There was no problem in the prenatal, natal and postnatal periods. It was learned that she was diagnosed with atypical autism. On examination, straight eyebrows, mild prognathism, cafe-au-lait spots were noticed on the trunk. Brain MRI and EEG were normal. The patient's hearing test was normal. It was planned to study karyotype and microarray from the patient. Karyotype result was 46,XX. As a result of the SNP-array, loss of heterozygosity (LOH) was detected in the 11p15.5p15.4 chromosomal region with a size of 9.4Mb and an estimated mosaicism of ~10%. As a result of the SNP-array study from the parents, the mosaic-UPD in the patient was found to be paternal UPD. With this result, the patient was diagnosed with BWS. Paternal UPD of chromosome 11 accounts for up to 20% of molecularly confirmed cases of BWSp. Upd(11)pat is usually the product of mitotic recombination errors and therefore appears as mosaic. It is therefore a challenge for diagnosis and can affect only parts of the body or organ. This disease is underdiagnosed because of low level mosaicism. In cases with normal results from blood, further examination with tissue biopsies is required. Upd(11)pat belongs to the BWSp subgroup with the second highest tumor risk and therefore requires special awareness in diagnosis and clinical management.

[OP-22]

Rare Case: Nablus Mask-Like Facial Syndrome

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Introduction

Nablus mask-like facial syndrome is a rare microdeletion syndrome associated with a distinctive facial appearance. The first case was identified in 2000 by Teebi, who observed a unique facial feature in a child from the Palestinian city of Nablus. Individuals with this syndrome typically exhibit features such as blepharophimosis, shiny and taut facial skin, sparse eyebrows, a bulbous nose, a protruding columella, and ear anomalies. Additionally, individuals with the syndrome may experience mental retardation and developmental delays. Nablus mask-like facial syndrome is exceptionally rare, with only 13 cases published to date. In this study, we will evaluate the genotype and phenotypic findings of our patient who presented to our clinic with speech delay and dysmorphic facial features and was subsequently diagnosed with Nablus mask-like facial syndrome.

Case Report

The patient is a 2.5-year-old boy born at 35 weeks of gestation. His parents are cousins. There were no prenatal or natal issues or concerns. There was no known occurrence of a similar condition within the family. It was learned that there was no similar patient in the family, sitting without support in the postnatal period occurred at 6-7 months, walking without support at 11 months and he could not speak yet. Body weight was 12 kg (10-25p) and height was 88 cm (10-25p). Physical examination revealed raised anterior hair, sparse eyebrows, bulbous nose, tense and shiny facial skin, microretrognathia, high palate, and recractile testis. Conventional karyotyping and molecular karyotyping with Affymetrix, CytoScan 315K (Affymetrix CytoScan Optima®) were performed on a peripheral blood sample obtained from our patient. The results were analyzed using the Chromosome Analysis Suite (ChAS) software. The conventional karyotyping result is 46XY, Molecular karyotyping showed a 1.5 MB deletion in the 8q22.1 region, spanning from 92,997,020 to 95,562,991, when compared to the GRCh38 reference genome.

Discussion

8q22.1 deletion syndrome, also recognized as Nablus mask-like facial syndrome, is an exceedingly rare condition. In our patient, the phenotypic characteristics and the 8q22.1 deletion identified through molecular karyotyping align with the features of Nablus mask-like facial syndrome. However, it is noteworthy to mention that there are documented cases in the literature where individuals with an 8q22.1 deletion exhibit mental retardation, developmental delays, and dysmorphic features, but they lack the typical facial findings of Nablus mask-like facial syndrome. Currently, the precise relationship between genotype and phenotype in this syndrome remains partially understood. Therefore, conducting a comparative analysis of the deletion region and the genes associated with similar cases may offer valuable insights into this matter.

[OP-23]

Analysis of Parental Chromosome and QF-PCR from Abortus Materials in our Genetic Diagnosis Center: Assessment of Results in a Retrospective Manner

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Abstract

Introduction: The aim of this study aims to determine as retrospectively analyze the results of the parental chromosomal analysis of the couples who pregnancy loss with QF-PCR analysis in patients who received abortion materials.

Methods: In this study, 1,237 parents who karyotyping analyzed and QF-PCR analysis of 624 abortus materials at the department Meram Medical Faculty Medical Genetics Laboratory of Necmettin Erbakan University were evaluated between 2014-2018 retrospectively.

Results: According to the results of our study; chromosomes anomaly was found in 11.8% of the QF-PCR analyzes. In these anomalies were 58.1% trisomy, 22.9% polyploidy, and 18.9% monozomy X. In family chromosome analysis findings, 97.1% of the parents have normal chromosomal organization, while 2.9% of them were abnormal. In the couples with chromosomal abnormalities; 0.6% variant, translocation carriers, 0.2% Turner syndrome, 0.2% Klinefelter syndrome (KS), 0.1% mosaicism, and 0.1% (Y) were detected.

Conclusion: Genetic anomalies are important in the etiology of abortion and the trisomies are the most common anomalies in our study as compatible with the literature studies. On the other hand 3 cases were encountered that is KS in tha data that have the the parental chromosome analysis. We believe that the results obtained make a significant contribution to current information will help to especially in the population preventive medicine applications and studies to be done for genetic counseling.

Keywords: Abortion, Klinefelter syndrome, prenatal diagnosis

[OP-24]

Clinical Exome Sequencing in Syndromic Epilepsy Patients Evaluation of Analysis Results

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Abstract

Introduction: Complex inheritance mostly plays a role in the development of epilepsy. In complex inheritance, the interaction of genes and environmental factors is seen. In our study, it was aimed to reveal clinical features of syndromic epilepsy patients, advantages-disadvantages of the clinical exome sequencing (CES), and diagnostic availability, to evaluate the relationship between the obtained molecular data and clinical features, and to provide genetic counseling to the families with the case whose molecular etiology was solved and the disease and variant.

Methods: A total of 146 patients, 70 men, and 76 women, who were diagnosed with epilepsy and had additional clinical findings such as syndromic appearance and mental retardation, were included in the study. Genes associated with syndromic epilepsy were analyzed using the CES kit containing 4,496 genes, using current databases with next generation sequencing (NGS) method.

Results: In our study, 17 (12%) pathogenic variants (P) with known clinical significance, 16 (11%) potentially pathogenic (LP) and 6 (3%) variant unknown significance (VUS) variants were detected in 38 (27%) individuals. Of the variants, 8 (5%) were in genes associated with encephalopathies, 6 (3%) were in genes associated with neurocutaneous diseases, 4 (2%) were in genes associated with metabolic pathways, and 21 (14%) variants are in genes that may cause rare syndromic epilepsy. In our study, 18 variants that can be classified as P, LP, or VUS were not reported in databases and were brought to the literature.

Conclusion: Genetic etiology was clarified and genotype-phenotype correlation was discussed in 38 patients with syndromic epilepsy. All these findings revealed the importance of NGS applications in elucidating the genetic mechanisms involved in the formation of epilepsy.

Keywords: Epilepsy, epilepsy genetics, clinical exome sequencing (CES)

[OP-25]

The Power of MLPA-NGS Coexistence in the Management of Charcot Marie Tooth Patients

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Abstract

Charcot Marie tooth (CMT) disease is an inherited motor and sensory neuropathies. Its prevalence is about 1/2500. Distal muscle weakness, muscle atrophy, loss of deep tendon reflexes, and foot deformities are expected findings. The findings are symmetrical, slowly progressive, and usually begin in the first decade. CMT can be inherited in an autosomal dominant, autosomal recessive, or X-linked manner. Duplications in the *PMP22* gene are responsible for the etiology in more than half of CMT patients. Nearly 80 genes are responsible for the other half, the most common being *PMP22*, *GJB1*, *MPZ*, *MFN2*, *GDAP1*, *HINT1*, *SH3CT2*, *SORD* genes. Eight patients aged between 3-55 years were admitted to our center with complaints such as difficulty in walking, electromyography abnormalities, loss of sensation in the hands and feet. MLPA analysis and next generation sequencing (NGS) analysis were performed for the etiology of the patients. Unlike the literature, no duplication in the *PMP22* gene was observed in our patients in MLPA; variants were detected in the gene panels of 4 of our patients. The variants are in the *GJB1*, *MPZ*, and *ARHGEF10* genes. The c.397A>C:I127L homozygous variant in *GJB1* gene detected in a 17-year-old male patient who presented with gait disturbance and motor and sensory polyneuropathy is novel. Although it is known that *PMP22* gene duplications are frequently responsible for the etiology of CMT, all of our patients had normal MLPA and found responsible variants by NGS method; it emphasizes the importance of the technical combination of MLPA and NGS in the approach to CMT patients.

Keywords: Charcot Marie tooth, MLPA, next generation sequencing

[OP-26]

Waardenburg Syndrome to PCWH Syndrome: Clinical Heterogeneity in the *SOX10* Gene

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Introduction

SOX10 is a transcription factor that plays a role in the differentiation of melanocyte vestibular and Schwann cells. Mutations in the *SOX10* gene cause Waardenburg syndrome, which includes hearing loss, iris heterochromia, and piebaldism. It can present with a broad clinical spectrum, from PCWH syndrome to which neurological findings such as polyneuropathy and Hirschprung's disease are also added. In our study, we presented the clinical heterogeneity shown by the pathogenic variant in the *SOX10* gene in the family.

Case Report

The patient who applied to our medical genetics outpatient clinic was a 6-year-old boy with a total colectomy due to Hirschprung disease. Their parents were 1st-degree cousins. On physical examination, satyr ears, bright blue tint, ptosis in the eyes, dental anomalies, and proportionate short stature were observed. In his neurological examination, deep tendon reflexes were weak, and muscle strength in his extremities was evaluated as 3/5. In addition, the patient had ataxia, anosmia, mental retardation, and bilateral sensorineural hearing loss (SNHL). Vestibular magnetic resonance imaging revealed posterior semicircular canal atresia, bilateral vestibular dysgenesis and vestibular aqueduct agenesis. Electromyography revealed sensorineural polyneuropathy with axonal destruction. Additionally, fundus examination revealed peripapillary atrophy and fundus albinous. The patient's father and brother had bilateral SNHL, iris heterochromia, anosmia, and large incisive teeth. Piebaldism was observed only in the father. As a result of clinical evaluations, *SOX10* whole gene analysis was planned with the preliminary diagnosis of index PCWH syndrome and paternal and maternal Waardenburg syndrome type 4C. *SOX10* whole gene sequence analysis was performed by the Sanger sequencing method. Bioinformatic analyses were classified in line with the "ACMG" guide. Detailed counseling was given before/after the test.

Conclusion

As a result of the analyses, a heterozygous p.Ser13Asnfs*19 (c.38_39delGCinsA) variant was detected in the index, father, and mother. Although this variant is a new variant that has not been previously reported in the literature, it was evaluated as pathogenic *in silico* analyses. It is thought that the mutation shows variable expressivity and clinical heterogeneity in the family and may contribute to the clinical evaluation of Waardenburg disease.

[OP-27]

A Family Diagnosed with Progressive Spastic Tetraplegia and Axial Hypotonia and Amyotrophic Lateral Sclerosis with a Pathogenic Variant in the *SOD1* Gene

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Introduction

Progressive spastic tetraplegia and axial hypotonia (PSTAH) is an autosomal recessive neurological disease characterized by the onset of severe and progressive motor dysfunction in the first year of life. Affected individuals have severe axial hypotonia with spastic tetraplegia and hyperekplexia reflecting upper motor neuron involvement. Pathogenic variants in the biallelic *SOD1* gene have been associated with PSTAH syndrome. Monoallelic pathogenic variants in the *SOD1* gene have been associated with ALS. In our study, we aimed to evaluate the connection between molecular data and clinical features and to provide genetic counseling regarding the disease and variant to the cases with determined molecular etiology and their families. After DNA isolation from the peripheral blood of the cases, genes associated with spasticity were analyzed using the gene-containing CES kit and the next generation sequencing method using current databases. Sanger sequencing method was used for confirmation and segregation analysis of the variants detected in the cases.

Case Report

A three-year-old boy whose parents were cousins was followed up due to hypotonia and loss of gross and fine motor functions. In the neurological examination of the patient, severe, progressive spastic tetraparesis with extensor plantar responses was observed. Some fasciculations were observed without other definitive electromyography signs of lower motor neuron involvement. His cognitive development was appropriate for his age, although he was non-verbal. Brain imaging was consistent with mild frontoparietal atrophy. In the genetic analysis of the case, a homozygous c.247dup p.(Asp84Argfs*8) possible pathogenic variant was detected in the *SOD1* gene. In the segregation analysis, the variant was detected as heterozygous in both parents. It was learned that the mother of the case was followed up with a preliminary diagnosis of ALS due to muscle weakness and fasciculation.

[OP-28]

Adaptor Protein Complex 4-associated Hereditary Spastic Paraplegia: A Case Series of Seven Patients

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Abstract

AP-4 is a heterotetrameric protein complex belonging to the adapter proteins (AP-1-AP-5) family, which facilitates the selective uptake of transmembrane cargo proteins into vesicles and assists their intracellular exchange. Biallelic pathogenic variants in one of four genes *(AP4B1, -E1, -M1, -S1*) encoding subunits of the AP-4 complex (β4, ε4, μ4, σ4, respectively) result in AP-4-associated hereditary spastic paraplegia (HSP). Its prevalence is estimated to be <1/1000000. In this study, we present a case series of 7 patients with AP-4-related HSP from 5 different families. Patients referred to our department due to spasticity between 2019-2022 were retrospectively reviewed. Seven cases diagnosed with AP-4-related HSP were included in the study. By clinical exome sequencing, homozygous mutations were detected in *AP4M1* in 6 patients and in *AP4B1* in 1 patient. The most common findings in the patients were spasticity (7/7), neurodevelopmental delay (7/7), microcephaly (6/7), and epilepsy (5/7). When we evaluated the variant spectrum, there were 3 different variants in the *AP4M1* gene. The c.1012C>T (p.Arg338*) variant, which is frequently mutated in Turkish population and thought to have a founder effect, was the most common change in our cohort (3/6). In addition, a homozygous c.975-25_975-22delGACT (rs767251230) variant was detected in a girl from a consanguineous Afghan family. This is a rare variant (gnomaAD frequency 0.0008%) that has not been previously reported in patients. In summary, we aim to contribute to the literature with 7 patients diagnosed with AP-4-related HSP, which constitutes a rare part of HSP, and we present the homozygous c.975-25_975-22delGACT variant in *AP4M1,* which has not been reported in any patient before. In addition, this study contributes to the claims that the c.1012C>T (p.Arg338*) variant, which is detected at a high rate despite the small number of patients, may have a founder effect in the Turkish population.

Keywords: Hereditary spastic paraplegia, *AP4M1*, *AP4B1*

[OP-29]

A Rare Syndrome with New Phenotypic Features and Mutation: Weiss Kruzska Syndrome

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Introduction

Weiss Kruzska syndrome is a rare disease accompanied by multiple congenital anomalies. It manifests with growth and developmental delays and branchiofacial anomalies. Affected individuals experience hypotonia, feeding problems, and corpus callosum agenesis, as observed in brain magnetic resonance images. The *ZNF462* gene encodes a transcription factor that plays a crucial role in embryonic development and chromatin remodeling. Pathogenic variants causing loss of function are implicated in the etiology of Weiss Kruzska syndrome, which is inherited in an autosomal dominant manner.

Case Report

A 5-year-old male patient was admitted to our outpatient clinic in the department of medical genetics due to growth and developmental retardation, frequent infections, and a dysmorphic facial appearance. The physical examination revealed skin tags in bilateral ears, pits in the right ear, fistula, pads in the fingers, an epidermoid cyst in the right eye, mane neck, pectus excavatum, and laryngomalacia. ECHO examination showed ASD, while laboratory tests indicated low immunoglobulin, and CD levels. Intravenous immunoglobulin treatment was initiated. Additionally, the patient's father had a similar history of immunodeficiency, and his dysmorphic features coincided with those of the patient.

Whole exome sequencing was performed using the next-generation sequencing method. Bioinformatic analyses were conducted following the "ACMG" guidelines via QCI-Analyse and QCI-Interpret. The analyses revealed a heterozygous p.R451*(c.1351C>T) variant in the *ZNF462* gene. This novel, previously unidentified variant was deemed pathogenic in *in silico* analyses. The variant detected in our index case was also examined in the parents, and the same variant was identified in the phenotypically similar father.

Conclusion

Immunodeficiency has not been identified to date in Weiss Kruzska syndrome, which includes developmental delays and non-specific dysmorphic findings. Its clinical features, overlapping with many syndromes, are quite heterogeneous. Usually diagnosed through whole exome sequencing, Weiss Kruzska syndrome is a rare condition that should be considered in the differential diagnosis.

[OP-30]

Coffin Siris Syndrome with Corpus Callosum Agenesia

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Introduction

Coffin-Siris syndrome (CSS) is a rare genetic disease with multisystem involvement characterized by intellectual disability. Fifth finger distal phalanx/nail aplasia or hypoplasia, developmental delay, facial dysmorphism, hypotonia, hirsutism/hypertrichosis and nervous system involvement are among the components of the CSS. We aim to make a genotype-phenotype comparison of the current clinical findings of CSS and to contribute to the literature with the new clinical findings detected in our patient.

Case Report

7-year-old female patient had corpus callosum agenesis, seizure, strabismus, learning difficulties, hyperlaxity, and cutis laxa. In the pedigree analysis there was a 3rd degree consanguinity between the parents and no similar history was found in the family. It was determined that the patient had a retardation in the developmental stages. Physical examination revealed facial dysmorphism and hypoplasia of the nails.

Results

Chromosome analysis from the patient's peripheral blood was found to be 46,XX and no deletion-duplication was detected in the microarray result. Molecular analysis of clinical exome sequencing method revealed a heterozygous c.5704A>T p.Lys1902* in the *ARID1B* gene (NM:001374828). This variant was not reported on Clinvar but according to ACMG criteria it was evaluated as likely pathogenic and segregation analyzed was planned.

Discussion

There are 12 genes related to CSS and the clinical findings are quite heterogeneous. So it is difficult to make genotype-phenotype correlation of CSS. In conclusion, this study reveals an unreported clinical finding in the literature and contributes to the genotype-phenotype correlation of CSS.

[OP-31]

A Rare Syndrome with a Rare Complication: Schimke Immunoosseous Dysplasia and Cerebral Hemorrhage

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Introduction

Schimke immunoosseous dysplasia (SIOD) is an autosomal ressesive disorder characterized by spondyloepiphyseal dysplasia, cellular immune deficiency, and nephropathy. Biallelic mutations in SWI/SNF-related matrix-associated actin-dependent regulator of chromatin subfamily A-like protein (*SMARCL1*) gene is the only gene responsible of the phenotype. The symptoms associated with cranium more likely present as ischemia, internal carotid artery atheroscleros and moyamoya phenomenon.

Case Report

Here, we describe a 20-months-old girl who admitted to hospital for failure to thrive. She was the only child of a consanguineous family. Intrauterine growth deficiency was noted. On examination her weight, height and occiputofrontal diameter were below -6 SD. She had large anterior fontanel, relative macrocephaly, bitemporal narrowing, thick eyelashes, prominent nasal tip, short trunk, and lumbar kyphosis. Although she had developmental delay she could not neither sit nor hold her head steady for the last one week. On X-ray, osteopenia, small capital femoral epiphyses, hypoplasia of the basilar portions of the ilia were detected. Brain imaging showed wide hypodensity on the left frontotemporal and right temporoparietal areas depicting parenchymal hemorrhage. *SMARCAL1*:c.1687C>T homozygous pathogenic variant was identified. During the follow-up, the patient's clinical condition deteriorated rapidly and deceased.

Conclusion

Reduced blood flow to the brain (cerebral ischemia) is one of the rare complications of SIOD. However, sudden death secondary to cerebral hemorrhage and ischemia in infancy has not been reported before. It should be kept in mind in regular neurological examination and cranial imaging should be performed from the early infancy.

[OP-32]

A Rare Case Report of Two Siblings: A Novel GRIN1 Variant

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Introduction

Neurodevelopmental disorder with or without hyperkinetic movements and seizures (NHDMS) (OMIM:138249) is a rare genetic disease characterized by psychomotor developmental delay, autism, and dysmorphic facial features. Monoallelic or biallelic mutations in the *GRIN1* gene which encodes NMDA channel subunit 1 have been associated with the NHDMS. Our aim is to present the clinical findings of two siblings with homozygous variants in the *GRIN1* gene and discuss the phenotype- genotype correlation.

Case Report

Two siblings, a 5-year-old girl and an 11-year-old boy were referred to our clinic due to global neurodevelopmental delay. Their parents are first-degree cousins, and there is no similar history in the family. In the detailed anamnesis and physical examination, the boy had hypotonia, autism, involuntary movements, speech delay, and self-injurious tendencies, and dysmorphic facial features. Also, the sister had dystonia, gait and speech delay, strabismus, and dysmorphic facial features. The siblings had normal results of metabolic tests, cranial magnetic resonance, abdominal ultrasound, electroencephalogram, and echocardiogram.

Results

Molecular analysis of the whole-exome sequencing method revealed a homozygous c.1640C>T (p.Pro547Leu) in the *GRIN1* gene (NM:007327). This variant was evaluated as likely pathogenic according to ACMG criteria. Segregation analysis revealed that the parents were heterozygous while the sister was homozygous for this mutation.

Discussion

The reporting of new cases is important for a better understanding of the characteristics of the rare genetic diseases. To conclude this study reveals a novel pathogenic variation to the literature and contributes to the phenotype-genotype correlation for NHDMS.

[OP-33]

Co-occurrence of Williams-Beuren Syndrome and Mosaic Turner Syndrome

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Introduction

Turner's syndrome (TS) is a form of gonadal dysgenesis with an incidence of 1:2000 to 1:5000 in live female births. The karyotype 45,X/46,XX is the most frequent mosaic type of this disease (36%). Williams-Beuren syndrome (WBS) is a microdeletion syndrome with a frequency of 1/20,000 caused by del7q11.23. Here, we report co-occurrence of WBS and mosaic TS, which has not been previously reported in the literature.

Case Report

Three years old female patient was admitted to our outpatient clinic with complaints of growth and neuromotor developmental retardation. She was born to a 22 year-old mother; her birth weight was 2700 g. On physical examination, her height was 96 cm (-1.18 SDS) and her weight was 14.8 kg (-0.46 SDS). The patient's findings include long philtrum, down-sloping shoulders, microdontia, hypertelorism, strabismus, large ears, thin upper lip, thick lower lip vermilion, wide nasal tip, anteverted nares, high forehead, wide mouth, and periorbital fullness. Laboratory examinations revealed hypothyroidism; and echocardiography showed supravalvular aortic stenosis. Karyotype and FISH analysis for WBS were planned in the light of the clinical findings. Chromosome analysis of lymphocytes showed mosaicism for TS (45,X[8]/46,XX[42]) and FISH analysis revealed del7q11.23 in all metaphases and interphases examined.

Conclusion

Co-occurrence of two distinct chromosomal abnormalities is a extremely rare phenomenon. In literature, there is only one report of co-occurring WBS and TS without mosaicism. This is the first report of a patient with both WBS and mosaic TS to our knowledge.

[OP-34]

PGT-M for Neurogenetic Diseases with Monogenic Inheritance: Applications and Experiences

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Abstract

Introduction: PGT-M can be recommended to all monogenic diseases where the disease causing loci are well defined. The most common indications for PGT-M are cystic fibrosis hemoglobinopathies for otosomal recessive disorders, myotonic dystrophy type 1, neurofibromatosis, Huntington's disease and hereditary cancer syndromes for autosomal dominant disorders. PGT for X linked disorders primarily performed for Duchenne muscular dystrophy (DMD), hemophilia, and fragile X syndrome.

Methods: This study discusses materials and methods of PGT-M studies for neurodevelopmental disorders performed in our department including DMD, spinal muscular atrophy, carbamoyl phosphate synthetase 1 deficiency, I-cell disease (mucolipidosis type 2), spastic paraplegia type 15, intellectual developmental disorder autosomal recessive 72, and asparagine synthetase deficiency. Families who sought to become pregnant by selecting healthy embryos and had identified pathogenic variants associated with single-gene diseases were included in this study.

Results: Twenty two trophectoderm embryo biopsies were sent to our institution for PGT-M studies. Whole genome amplification could not be achieved 7 out of the 22 embryos. Among the 15 embryos that were evaluated, 4 were found to be wild-type, 4 were carriers, 3 were homozygous mutants, and 1 was hemizygous. Four clinical pregnancy observed after 8 embryo were transferred from 6 different families.

Discussion: The choice of method in PGT studies can vary depending on the inheritance pattern of diseases and mutation types. The reason for application may differ, such as selecting healthy embryos or HLA-compatible embryos. During the PGT-M study process, families seeking to have children should be informed about potential situations that may arise (failed implantation, multiple pregnancies, inability to find a transferrable embryo) through genetic counseling. Technical and clinical experiences during this study will serve as a guide for future studies.

[OP-35]

Blended Phenotype in a Case with Brain Malformation, Neurodevelopmental Disorder and Epilepsy

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Abstract

One of the important causes of severe neuromotor developmental delay and epilepsy is malformations of cortical development (MCD) such as pachygyria, lissencephaly. Genes involved in neuronal cell proliferation, migration and post-migration cortical organization have been implicated in MCD. The *CAMPAS1* gene, which provides microtubule and spectrin binding activity, is one of the rare causes that have been recently identified. Another rare cause of severe neuromotor developmental delay and epilepsy is he *NBEA* gene. It encodes the neurobeachin protein, a neuro-specific structural protein that plays a role in vesicle traffic and synaptic structure. In this study, we present a case with loss of function variant in these two rare genes. A proband 7-year-old female patient, only child of consanguineous parents, microcephaly, seizure, infantile spasm, neurodevelopmental retardation, hypotonia, pachygyria, corpus callosum agenesis, hypoplasia of the brain stem, spasticity in the extremities, nutrition problems, recurrent respiratory tract infection findings. Whole exome analysis of the patient revealed c.1153C>T p.Gln385* non-sense homozygous likely pathogenic pathogenic variant in *CAMSAP1* gene and *de novo* c.6867G>A p.Trp2289* non-sense heterozygous pathogenic variant in *NBEA* gene. We considered it as a blended phenotype. In the literature, the case of these two genes together has not been reported before. There are limited case reports with these two genes. For this reason, it is thought that it will contribute to the literature. In this study, we emphasize that in the presence of complex and severe clinical findings, two or more genes may be responsible and further investigation may be required.

[OP-36]

Evaluation of Genetic Disorders Associated with Epilepsy in Children

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Abstract

Introduction: Epilepsy is a neurological disorder characterized by recurrent seizures due to increased neuronal and electrical activity. According to the World Health Organization, approximately 5 million people are diagnosed with epilepsy for the first time each year. While idiopathic epilepsy has no identifiable underlying cause, genetic factors play a role in many cases. Advances in genetic science have led to the identification of many genetic syndromes and single-gene mutations associated with epilepsy. Many syndromes associated with or caused by chromosomal aneuploidies, copy number variations, and single-gene mutations have been reported in association with epilepsy. This study aimed to evaluate the genetic syndromes detected in patients with epilepsy.

Methods: We retrospectively analyzed the genetic test results of 230 patients with epilepsy who presented to our department between January 2016 and January 2023.

Results: Abnormalities were detected in 30 patients (13%). Chromosomal anomalies were found in four patients, microdeletion syndrome in 14 patients, and single-gene mutations in 12 patients. The most common single-gene mutations were in the *ADGRV1, ARSA, ATN1, COL4A1, CPA6, KCNQ, MACF1, PACS1, PIGQ, PRRT2, SLC19A2,* and *STXBP1* genes.

Conclusion: Genetic factors play a significant role in the development of epilepsy. Genetic testing can be helpful in identifying the underlying genetic cause in syndromic and non-syndromic cases, which can aid in diagnosis, prognosis, and counseling.

Keywords: Epilepsy, genetics, diagnosis

[OP-37]

A Case of 48,XXYY Syndrome Presenting with ADHD

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Introduction

Attention deficit hyperactivity disorder (ADHD) which is characterized by increased movement, decreased impulsivity control and attention span is the most common neurobehavioral condition in childhood (1). Research suggests that ADHD is one of the psychiatric disorders with the highest association with genetic abnormalities. Mutations in genes such as *5HTT, DAT1, DRD4, DRD5, HTR1B, SNAP25, BAIAP2, SLC6A3* have been implicated as well as copy number changes (2). Determining the causes related to chromosomal abnormalities in neurodevelopmental disorders such as ADHD, which exhibit genetic heterogeneity, is important in terms of clinical approach and assessing genetic risks. 48,XXYY syndrome is a syndrome that occurs in one out of every 18,000-50,000 male infants and was previously classified as a variant of Klinefelter syndrome. However, as more cases were reported, its unique psychiatric spectrum and set of findings led the medical community to recognize 48,XXYY syndrome as a separate condition. It is characterized by delayed speech and motor skills, hypotonia and feeding problems. Additionally, epilepsy (15%), tremors (60%), asthma and allergies (60%), and dental problems (90%) can also be present (3). ADHD, among other psychiatric conditions, has also been reported by some studies (4).

Case Report

We present a 7-year-old male patient who was referred to pediatric neurology for difficulty in learning and behavioral issues. There was no family history of any mental illness or consanguinity. He was later diagnosed with ADHD. His physical examination was normal except for mild hypertelorism. He had a history of cryptorchidism. He also had a history of epilepsy and was seizure-free with anti-epileptics. His magnetic resonance imaging was normal except for non-specific T_2 hyperintensities in the white matter. Karyotyping was performed from peripheral blood and 48,XXYY was recognized on all thirty metaphases analyzed.

Discussion

Neuropsychiatric disorders are multifactorial with a strong genetic component. This genetic component is usually on a molecular level (5). However, if there are certain dysmorphic findings and no family history, starting the genetic workup with a basic karyotype can be both timesaving and cost-

effective. Finally, due to its multiple long-term complications (neuropsychiatric problems, hypogonadism, inguinal hernias, vascular conditions, dental issues, asthma, type 2 diabetes, congenital cardiac anomalies, etc.), 48,XXYY syndrome should be considered as a distinct clinical and genetic entity and not just a variant of Klinefelter syndrome. For this distinction, it is important to provide genetic counseling to the family and create awareness for their future medical care needs.

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[OP-38]

A Rare Variant in the *CDH2* Gene: The Second ACOGS Case from Türkiye

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Introduction

Agenesis of the corpus callosum, cardiac, ocular, and genital syndrome (ACOGS) (OMIM: 618929) is a rare genetic disorder caused by heterozygous mutations in the *CDH2* gene. The *CDH2* gene located at the 18q12 locus encodes N (neuronal)-cadherin, which plays a crucial role in early brain morphogenesis. ACOGS is a neurodevelopmental disorder characterized by developmental delay and/or intellectual disability, corpus callosum agenesis/hypoplasia, craniofacial dysmorphisms, and ocular, cardiac, and genital anomalies.

Case Report

A 2-year-old male patient was genetically evaluated due to craniofacial dysmorphic features, strabismus, limited upward gaze, epilepsy, intellectual disability, speech delay, sexual development disorder, and cranial magnetic resonance findings of corpus callosum agenesis and scaphocephaly. The patient's echocardiogram showed patent ductus arteriosus, patent foramen ovale, left ventricular non-compaction. A cystoscopy revealed scrotal hypoplasia, hypoplastic phallus, absence of the right gonad, and atrophic testicular structure on the left. Whole exome sequencing analysis identified a heterozygous intronic variant in the *CDH2* gene (c.1344+5G>A).

Conclusion

Although this variant in the *CDH2* gene is listed in the ClinVar database, it has not been reported in the literature before. Additionally, this is the second reported ACOGS case from Türkiye. The discovery of non-compaction cardiomyopathy in our patient may represent a new phenotype associated with ACOGS.

[OP-39]

An Interesting Family: A Patient with Blended Phenotype with Sexual Development Disorder and Coenzyme Q10 Deficiency and His Sibling Diagnosed with Joubert

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Abstract

Consanguineous marriages cause rare blended phenotypes in common geographies and the emergence of different homozygous diseases in the same family. In this article, a family in which different rare mechanisms come together as a result of consanguineous marriage is presented. A 2-year-old proband with a blended phenotype with sex development disorder and coenzyme Q10 deficiency has psychomotor retardation, micropenis,

undescended testicles, dysmorphic findings, hypotonia, and the gonads have a bilateral testicular appearance and are located in the inguinal canal. In the patient's brother, who previously died due to hydrocephalus, a non-sense homozygous variant c.1051C>T p.(Arg351*) was detected in the 7th exon of the NM_001134830 transcript of the *AHI1* gene. The patient's cytogenetic analysis result is: 46,XX,ish,der(X)t(X;Y)(p22.3;p11.2)(SRY+). In the molecular karyotyping analysis performed due to hypotonia, psychomotor retardation and dysmorphic findings; a change of 2.1 Mb in size was detected in the Xp22.33p22.32 region and 3.1 Mb in size in the Yp11.2 region. This change is 46,XX sex reversal 1; it has been associated with SRXX1 (OMIM number:#400045) disease. In the subsequent whole exome analysis, a c.437T>G (Phe146Cys) missense homozygous probable pathogenic variant was detected in the NM_016035 transcript in the 5th exon of the *COQ4* gene, which explains other clinical findings. In primary deficiency of coenzyme Q10 (OMIM number: #616276), which is a lipid component of the mitochondrial respiratory chain, symptoms vary; hypotonia, decreased coQ levels in muscle tissue, regression in psychomotor development, seizures, and increased serum lactate levels are observed. It is generally lethal in the first years of life, and in this respect, it is thought that the clinical findings of the proband will contribute to the literature with their milder course. To our knowledge, there is no other case in the literature reporting a sexual development anomaly or primary CoQ10 deficiency.

Keywords: SRY, gender development disorder, hypotonia, coenzyme Q10, COQ4

[OP-40]

Diagnosis of Townes-Brocks Syndrome in a Turkish Adolescent with End-Stage Renal Failure

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Abstract

Introduction: Townes-Brocks syndrome (TBS) is identified by a triad of imperforate anus, malformed ears, and thumb abnormalities. Additional associated features include renal complications, congenital heart abnormalities, foot abnormalities, and genitourinary malformations, and intellectual disability. Less common characteristics encompass iris coloboma, Duane anomaly, Arnold-Chiari malformation type 1, and developmental delay. Here we report a 15-year-old male patient with congenital hearing loss, global developmental delay, and the incidental discovery of chronic kidney disease. The patient's physical examination indicated TBS. However, clinical exon sequencing (CES) genetic testing revealed a pathogenic mutation in the *SALL1* gene and confirmed the diagnosis.

Methods: We took the patient for clinical exome sequencing.

Results: CES analysis revealed a novel heterozygous likely pathogenic variant in *SALL1* gene (NM_002968/3 c.1336_1370del p.(Phe446Leufs4)). We also detected a hemizygous likely pathogenic variant in the *POU3F4* gene (NM_000307/5 c.478C>T p.(Gln160)), which could similarly lead to the patient's congenital deafness (OMIM #304400 Deafness, X-linked 2). This suspicion arose because the patient's cousin also has congenital hearing loss without any other anomalies. Thus, we have arranged for further genetic examination of the cousin.

Conclusion: Our patient had a frameshift mutation in the hotspot region, leading to premature truncation. This resulted in a severe TBS phenotype with severe dental issues, which led to the extraction of teeth during childhood due to significant mouth deformities and functional problems. Recent studies have also emphasized that premature *SALL1* protein truncation results in a more severe TBS phenotype due to a dominant negative effect.

[OP-41]

Detection of Somatic Variant in *PIK3R2* Gene in a Patient Followed with Galactosemia

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Introduction

Megalencephaly-polymicrogyria-polydactyly-hydrocephalus (MPPH) syndrome is a rare developmental brain disorder characterized by brain enlargement, bilateral perisylvian polymicrogyria (BPP) and variable ventriculomegaly, and occurs as a result of pathogenic mutations in the *AKT3, CCND2 or PIK3R2* genes. Variable degrees of ventriculomegaly are observed in almost all children with MPPH syndrome. In this case report, we present a case referred to our clinic due to hemimagalencephaly, BPP and galactosemia. In the patient we underwent clinical exome sequencing (CES), we detected possible pathogenic and pathogenic variants in the *PIK3R2* gene (20% variant fraction) and *GALT* gene, respectively. The *PIK3R2*

gene regulates the activity of an enzyme called phosphatidylinositol 3-kinase (PI3K). The PI3K-AKT-mTOR pathway is essential for the normal development of many parts of the body, including the brain. Mutations in the *PIK3R2* gene can cause excessive activation of the PI3K/Akt/mTOR pathway, resulting in overgrowth of neurons to form developmental brain disorder syndrome, both MPPH and BPP, characterized by seizures and other clinical features.

Galactosemia is an autosomal recessive inherited condition that affects galactose metabolism, and is caused by the deficiency of the enzyme galactose-1-phosphate uridyltransferase (GALT). Classical galactosemia is divided into three types: Clinical variant galactosemia and biochemical variant galactosemia, the most common and most severe form being the classical one.

Case Report

In this study, we present a case diagnosed with galactosemia, which we associate with hemimagalencephaly and BPP. Our case is a 4-year-old girl patient. As a result of CES performed on the patient, c.329-2A>C splice acceptor, pathogenic variant was detected in the *GALT* gene and c.1669G>T p.(Asp557Tyr) possible pathogenic variant was detected in the *PIK3R2* gene (20% variant fraction).

Result

The variants detected in the patient were evaluated as blend phenotype. This study contributed to the importance of next-generation sequencing in detecting somatic mutations. It should be noted that somatic variants may not be of blood origin, so analysis of a second tissue such as saliva or skin should be considered. It should be noted that somatic variants cannot be detected in blood, so analysis of a second tissue such as saliva or skin should be considered.

Keywords: Galactosemia, *GALT,* BPP, *PIK3R2,* hemimegalencephaly

[OP-42]

A Novel Homozygous Variant in *TBC1D24* Gene: A Case Report

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Introduction

DOORS syndrome (deafness, onychodystrophy, osteodystrophy, mental retardation and seizures; MIM 220500), caused by variants in *TBC1D24* gene (TBC1 domain family member 24, MIM 613577), is a rare autosomal recessive genetic neurometabolic disorder. DOORS syndrome has five universal and prominent symptoms, which included deafness, absent or hypoplastic finger and/or toenails, finger deformities, mental retardation, and seizures. In addition, ophthalmologic anomalies, peripheral neuropathy, and characteristic coarse facial features such as a large nose with a wide nasal bridge, bulbous tip and anteverted nares, a long prominent philtrum, and downturned corners of the mouth can be observed. Less common findings included dental, kidney, and cardiac anomalies.

Case Report

A 1-year-old male patient, the child of a non-consanguineous couple, was genetically evaluated with infantile resistant epilepsy, hypotonia, nail dystrophy, digit anomaly, hearing loss, and dysmorphic facial findings. Karyotype and microarray analysis were 46,XY. Whole exome sequencing revealed a homozygous missense c.463G>C (Ala155Pro) variant in *TBC1D24* gene.

Discussion

In this study, we report a novel variants in DOORS syndrome and contribute to the clinical and molecular spectrum of the disorder.

[OP-43]

Deletion of Multiple Exons of KIF1C Detected by Next Generation Sequencing Associated with Spastic Ataxia 2

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Abstract

Introduction: Spastic ataxia 2 (SPAX2) is a rare, complex subtype of hereditary spastic paraplegia characterized by onset in the first two decades of cerebellar ataxia, associated with gait disturbances, dysarthria, and variable spasticity of the lower limbs. Autosomal recessive SPAX2 is caused by homozygous mutation in the *KIF1C* gene on chromosome 17p13. In 2014 Novarino et al. identified a homozygous deletion of exons 14-18 of the *KIF1C* gene in patients with SPAX2.

Methods: A 15-year-old male was referred with frequent falls, tremor, dysarthria, lower limb spasticity, hyperreflexia and specific learning disability. Echocardiography showed patent foramen ovale but no abnormal findings were found by brain magnetic resonance imaging, magnetic resonance spectroscopy, electroencephalography, electromyography and metabolic tests. In a NGS panel of 63 genes targeted for hereditary spastic paraplegia, all exonic regions and exon-intron junctions were analyzed by Qiagen Clinical Insight Interpret software after DNA isolation from peripheral blood of the patient. Microarray analyzes were performed with Illumina iScan System, 700K.

Results: No pathogenic mutation was detected in targeted NGS panel analysis, but homozygous deletion was suspected because exons 19-23 of the *KIF1C* gene could not be sequenced. Microarray analysis revealed a ~14-kb homozygous deletion of *KIF1C* and *SLC52A1* (arr[GRCh38]1 7p13.2(5020462_5034482)x0).

Conclusion: To the best of our knowledge, there is no previous report of SPAX2 with deletion of exons 19-23 of the *KIF1C* gene. We aimed to contribute to the literature about this rare form of spastic ataxia with the findings of this patient.

[OP-44]

Confirmation of *PATL1* Gene as Neurodevelopmental Disease Gene by Fruit Fly Model

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Abstract

Neurodevelopmental disorders are characterized by limitations in social behavior and intellectual disability (ID). To understand the genetic causes of ID and examine the biological pathways underlying ID risk genes reported by next-generation sequencing studies, the fruit fly *Drosophila melanogaster* is an ideal model, allowing screening of candidate genes with simple analyses. *PATL1* gene variants detected in two independent patients diagnosed with ID provided strong evidence that the *PATL1* gene is a candidate for ID. PATL1 protein localizes in cytoplasmic p bodies and regulates mRNA metabolism. *In vivo* studies revealed that when mutated, HPAT, the PATL1 ortholog in the fruit fly, causes synaptic defects at the neuromuscular junctions, and HPAT is vital for plasticity at synapses (Pradhan et al., 2012). We aimed to determine the genetic and neuromorphological consequences of the variations in the *PATL1* gene by performing functional and morphological analyses with mutant lines of the *HPAT* gene. We showed that knock-down of HPAT in the whole body causes lethality, and pan-neuronal knockdown of HPAT causes partial lethality with severe defects in motor functions. As a result of morphological analysis, we showed that HPAT is required for axon adhesion and guidance in the mushroom body in the fly brain, which is accepted as the analog of the human hippocampus. Also, we showed that HPAT has a critical role in the laminar organization of the ellipsoid body, the analog of the human brain basal ganglia in the fly brain central complex, which governs target-oriented navigation and motor control. All our findings highlight the critical role of HPAT in the central nervous system and synaptic functioning. This work is supported by TÜBİTAK project number: 122Z319.

Keywords: Neurodevelopmental disease, animal models, *Drosophila melanogaster,* p bodies, *PATL1*

[OP-45]

Insensitivity to Pain, Congenital, with Anhidrosis (CIPA): Presentation of 2 Siblings

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Introduction

Hereditary Sensory and Autonomic Neuropathy (HSAN) is a rare syndrome characterized by genetic heterogeneity. Insensitivity to pain and autonomic dysfunction are common features in affected patients. Homozygous or compound heterozygous mutations in the *NTRK1* gene have been associated with HSAN type 4 (CIPA), which is accompanied by anhidrosis among the common symptoms. The aim of our study is to discuss the clinical characteristics of two siblings in whom we detected mutations in the *NTRK1* gene within the context of genotype-phenotype relationship.

Case Report

Two Syrian-origin sisters were referred to our clinic due to recurrent fever episodes, anhidrosis, pain and temperature insensitivity, delayed wound healing, and self-mutilating behaviours. There were no specific prenatal, natal, or postnatal history features for the patients. However, both siblings had been hospitalized multiple times due to recurrent fever episodes. The siblings, whose parents had a first-cousin marriage, did not have a similar history in their family. During the physical examinations of the patients, common findings included bite marks on the tongue, amputations in the distal phalanges of their fingers due to self-biting, and non-healing wounds on their bodies. In the Whole Exome Sequencing analysis of the older sibling, a homozygous c.200delA (p.N67Tfs*2) frameshift variant in the *NTRK1* (NM_002529.3) gene was detected, which is not defined in ClinVar but evaluated as a potential pathogenic variant according to ACMG criteria. Segregation analysis in family members identified the parents as heterozygous carriers of the same pathogenic variant, while the affected sibling was homozygous for it.

Conclusion

Despite the rarity of CIPA as a genetic disease, a molecular diagnosis plays a significant role in screening at-risk individuals within the family, as well as in providing appropriate genetic counselling, conducting prenatal diagnosis, and following up with patients.

[OP-46]

A Case of Char Syndrome with a Novel TFAP2B Variant

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Introduction

Char syndrome (OMIM:169100) is a rare disorder characterized by dysmorphic facial features, patent ductus arteriosus, and hand anomalies. Its prevalence is unknown. The most common facial features are depressed nasal bridge, broad nasal tip, downslanted palpebral fissures, hypertelorism, short philtrum, and triangular mouth. The characteristic hand anomaly is aplasia or hypoplasia of the middle phalanges of the fifth fingers. Other congenital heart and extremity anomalies are also encountered. More rarely craniosynostosis, short stature, and other findings affecting organs such as teeth, eyes, and ears can also be seen. This autosomal dominantly inherited syndrome is caused by heterozygous disease-causing variants in *TFAP2B*. *TFAP2B* belongs to the *AP-2* gene family and encodes the transcription factor AP-2 beta. It is thought to have functions in cell proliferation and embryonic development.

Case Report

A 4-month-old female patient was consulted to our department with dysmorphic facial features and craniosynostosis. Hypertelorism, depressed nasal bridge, upslanted palpebral fissures, hypoplasia of the middle phalanges of the fifth fingers, and metopic synostosis were detected in the clinical examination. An echocardiogram revealed patent ductus arteriosus. Molecular testing with clinical exome sequencing revealed a novel heterozygous frameshift variant c.142del p.(Gln48Lysfs*48) in exon 2 of *TFAP2B* (NM_003221).

Discussion

Both haploinsufficiency and dominant-negative mechanisms have been reported in this syndrome, our case is likely caused by haploinsufficiency. Our report broadens the mutational spectrum of *TFAP2B* gene causing Char syndrome and highlights the importance of detailed physical examination and molecular testing in diagnosing rare syndromes.

[OP-47]

The Impact of Musical Performance on Cognitive Functions and Its Relationship with BDNF val66met and COMT val158met Polymorphisms

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Introduction: Musical training alters cognitive skills. The genetic contribution to these skills remained less explored. *BDNF* and *COMT* genes are related to brain plasticity. Drawing on a previous studies suggested that brain plasticity may be higher among musician we aimed to investigate the effect of musical training by using some tasks, which evaluate visuospatial and verbal skills, along with genetic profiling of *BDNF* and *COMT* polymorphisms.

Methods: Participants were 30 musicians and 30 control subjects with right-hand dominance. Cognitive assessments were done using Corsi block tapping (CBT), mental rotation (MRT), and verbal fluency tasks (VFT). CBT and MRT are visuospatial tasks performed separately by both hands. VFT has phonetic and semantic components. Genotype samples were taken from the participants using buccal brushes.

Results: CBT and VFT showed significant differences between the groups. Left-hand scores of the CBT were higher in musicians. Results show a positive change in the visuospatial abilities of musicians while using the non-dominant hand, which suggests that there is a relation between motor skills and visuospatial abilities. In the VFT, a significant decrease in errors made by musicians was found. Fewer errors made by musicians indicate that executive inhibition in musicians is better. Genotypes showed differences in some cognitive scores. Based on the results of our study, environmental variables, such as musical training, might be interacting with the *COMT* val158met polymorphism, specifically val allele, and thus give rise to differing cognitive phenotypes.

Conclusion: The study supports the idea that gene-environment interaction should be assessed together. Indicated polymorphisms seem to be candidate genes for determining the genetic mechanism.

[OP-48]

Examining the Effect of Sugar Diet on miR-126-3p Expression Level in Autistic Mouse Model

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Abstract

Autism is a multifactorial disorder that affects the central nervous system, with an etiology that has not been fully elucidated. Epigenetics refers to the study of changes that can directly affect gene expression without any alterations in DNA sequence. Epigenetic mechanisms can bring about changes in a gene's activity at transcriptional, post-transcriptional, translational, and post-translational levels. MicroRNAs (miRNAs), a subset of small non-coding RNAs, are among the best-characterized epigenetic mechanisms. MiRNAs play critical roles in processes such as development, protein secretion, and gene regulation. Studies in the literature have shown that different diet programs applied to autism patients can lead to changes in the severity of their behaviors. Additionally, diets used in various diseases have been shown to alter the expression levels of related genes. According to information from the literature, it has been found that the expression levels of six miRNAs are low in children with autism and their families. Therefore, valproic acid (VPA) mouse models, which exhibited autism-like behavior (confirmed through behavioral and molecular tests) were used. In the current study, newborn mice were fed diets containing 20%, 30%, and 10% (normal diet) sugar immediately after birth, and behavioral tests were conducted when they reached 2 months of age. The effects on the expression levels of autism-related marker miR-126-3p in the hippocampus, sperm, and blood tissues were examined. The results of our study showed significant differences in the hippocampus tissue of the VPA group compared to the group fed a 20% sugar diet with saline injection in behavioral experiments. Furthermore, there was a significant increase in sperm tissue in the group fed a 30% sugar diet compared to the control group. Our study results indicate that diets applied in autism can potentially alter both behavioral and molecular phenotypes. Additionally, the changes in sperm miRNA expression profiles due to sugar diet suggest that dietary habits may significantly impact subsequent generations.

Keyword: Autism, VPA, sugar diet, miR-126-3p

[OP-49]

Investigation of the Effects of Human REG3A Antimicrobial Protein on CD4+ T and Dendritic Cells *Ex Vivo*

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Introduction: Regenerative islet-derived protein 3-alpha (REG3A), also called heptocarcinoma-intestinal-pancreas or pancreas-associated protein, functions as an antimicrobial protein in the intestine. In this study, we investigated the immunomodulatory effects of REG3A, a member of the Reg family, on primary T-cells such with respect to polarization, cytokine production, proliferation, and on dendritic cell (DC) maturation.

Methods: Proliferation, apoptosis and IL-2 production by PBMCs obtained from blood taken from healthy volunteers using the Ficoll-Hypaque density gradient were examined. Naive CD4+ human T-cells were differentiated into selected Th1, Th2, Th17 and T-cells. CD14+ monocytes were differentiated into DCs. Select doses of recombinant human REG3A proteins were added to the culture. Flow cytometric analysis results were examined using FlowJo and Graphpad 9.

Discussion: Picogram doses of REG3A on T-cells did not cause statistically significant difference in terms of proliferation, apoptosis and IL-2 production. A dose-dependent decrease in FOXP3+ T-cells was seen but was not significant. REG3A exposure did not result in a significant difference in the expression of checkpoint inhibitors such as KLRG, PD-1, and CTLA4 by FOXP3+ T-cells. On the other hand, REG3A reduced Th17 cell-derived IL-17 and IL-22 production. Apoptosis experiments have shown that this reduction was due to cytotoxic effect of REG3A on Th17 cell cultures. Th2 cell-derived IL-4 production was not affected by REG3A exposure. When DCs were examined, REG3A exposure increased the surface expressions of ICOS and MHCII by mature DCs.

Conclusion: These results indicate that human REG3A has more pronounced effects on the Th17 lineage and may affect DC maturation. This project is part of thesis at ERÜ Health Sciences Institute, Department of Medical Biology a supported by the ERÜ BAP project with code TDK-2020 10779.

Keywords: Antigen presenting cell, CD4+ T-cell, REG3A

[OP-50]

Investigation of the Effect of Schisandrin B on Cytokine and Chemokine Production in Human Peripheral Blood Cells

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Introduction: The aim of this study is to characterize the effects of different doses of Sch-B on human immune cells, particularly T-cell subtypes, through *ex vivo* experiments at the molecular level.

Methods: To characterize the effects of Sch-B on human immune cells, lymphocytes isolated from peripheral blood of healthy donors were cultured with different doses of Sch-B for 24 hours, and the released chemokines and cytokines were evaluated using multiplex ELISA (the experiment was performed in quadruplicate). Lymphocytes cultured in Sch-B medium were stimulated with CD3/CD28 and curdlan. After stimulation biolegend multiplex ELISA was performed, followed by analysis and quantification using flow cytometry.

Results: Depending on the dose of Sch-B, there was a significant decrease in cytokine levels of interleukin (IL)-5, IL-13, IL-2, IL-6, IL-9, IL-10, and interferon-γ, while the levels of tumor necrosis factor-α, IL-17A, IL-17F, IL-4, and IL-22 cytokines significantly increased. Additionally, under conditions of Curdlan stimulation, there was a significant increase in IL-8 chemokine release with dose-dependent Sch-B, while the release of MIP-1α, MCP-1, ENA-78, and GRO-α chemokines showed a significant decrease. There were no significant changes in the release of IP-10, TARC, RANTES, MIP-3α, and MIP-1β chemokines between groups. In CD3/CD28-stimulated groups, there was a significant increase in IP-10, TARC, RANTES, and MIG chemokines, a significant decrease in MIP-1α, MCP-1, MIP-1β, ENA-78, MIP-3α, and GRO-α chemokines, while there was no change in IL-8 chemokine release.

Conclusion: Sch-B exhibits immunomodulatory properties, affecting the cytokines and chemokines released from human lymphocytes under *in vitro* conditions.

Keywords: Multiple sclerosis, lymphocytes, Th17 cell, treg cell, cell culture, multiplex ELISA

[OP-51]

Evaluation of the Effectiveness of Combined Treatment with the PD-L1 Inhibitor Atezolizumab and the c-MET Inhibitor Crizotinib in MCF-7 Breast Cancer Cells

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Abstract

Introduction: Atezolizumab is a PD-L1 inhibitor. PD-L1 immune checkpoint inhibitors are used in the treatment of breast cancer. However, response rates as monotherapy are limited. Additionally, most patients are resistant to treatment. c-MET has been reported to be associated with drug resistance in various cancer cells. Additionally, in studies showing the relationship between c-MET and immunotherapy, it has been reported that c-MET inhibitors increase the effectiveness of immunotherapies. Therefore, in our study, we evaluated the possible effectiveness of atezolizumab and c-MET inhibitor crizotinib combined treatment in MCF-7 breast cancer cells.

Methods: Some MCF-7 breast cancer cells were treated with atezolizumab, some cells were treated with crizotinib, and some cells were treated with atezolizumab + crizotinib. Additionally, some breast cancer cells were co-cultured with peripheral mononuclear blood cells (PMBC). Then, some of co-cultured cells were treated with atezolizumab, some with crizotinib, and some with atezolizumab + crizotinib. Then, cell viability (MTS analysis), colony formation (clonogenic detection) and apoptosis rates (annexin V analysis) of all these cells were analyzed.

Results: According to the results, it was found that atezolizumab had no effect on cell viability, colony formation and apoptosis rate in MCF-7 cells in the presence and absence of PMBC cells. It was found that crizotinib dose-dependently decreased cell viability and colony formation in MCF-7 cells and significantly increased the rate of apoptosis. It was found that the combination of atezolizumab + crizotinib in the presence of PMBC cells reduced cell viability and colony formation and increased the rate of apoptosis of cancer cells. However, it was found that these effects were similar to those on cells treated with crizotinib alone, so combined application did not increase the effectiveness of atezolizumab.

Conclusion: These findings show that atezolizumab has no effect on MCF-7 cells and crizotinib has a cytotoxic effect on breast cancer cells.

Keywords: Breast cancer, atezolizumab, PD-L1, crizotinib, c-MET

[OP-52]

A Metabolic Disease in which Neurological Findings are Prominent: Arginase Deficiency

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Abstract

Arginase deficiency (MIM#207800) is an autosomal recessive disease caused by compound heterozygous and homozygous mutations in the *ARG1* gene. It is an inborn error of metabolism resulting from a defect in the hydrolysis of arginine into urea and ornithine, in the last step of the urea cycle. It is characterized by episodic hyperammonemia, spasticity, loss of ambulation, seizures and severe intellectual disability in untreated individuals. Rarely, it can be life-threatening and cause death. A 10-year-old male patient was referred to us because of walking disorder. The patient's parents were sixth degree relatives. His siblings were healthy, but his mother's cousin's son also had difficulty walking. On physical examination, the patient had spasticity and ataxic gait. The patient had gait disorder for 5 years. Additionally, the patient had hypothyroidism, learning disability, and abnormal EEG findings. For these reasons, chromosome analysis and molecular karyotyping tests performed on the patient were normal. Also, when the *SPAST* and *ATP7B* gene analysis and the triplet repeats of *FXN* gene were found to be normal, whole exome sequencing (WES) test was performed on the patient. In the WES analysis, the c.702_703insAGACT (p.G235fs*20) variant in *ARG1* gene (NM_000045.4), which explains the patient's clinic, was detected as homozygous. This variant is a likely pathogenic variant causing loss of function. Comprehensive analysis such as WES analysis enable accurate diagnosis and appropriate clinical management. We also thought that we could contribute to the literature by presenting a new case about this rare disease.

[OP-53]

Relationship between *GSTP1, XRCC1, ERCC1, MTHFR TSER* and *DPYD* Gene Polymorphisms and Progression-Free Survival in Colorectal Cancer Patients Received FOLFOX Treatment

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Abstract

Introduction: In this study, we compared the genetic polymorphisms associated with 5-fluorourasil and oxaliplatin chemotherapy drugs and the progression-free survival times of stage IV and metastatic colon cancer patients who were treated only with the FOLFOX chemotherapy regimen. In this retrospective study, national and international ethical rules were complied with. This study approved Pamukkale University Ethic Committee with number of E-60116787-020-443991.

Methods: GSTP1-313A>G, XRCC1-G28152A, ERCC1-8092C>A, ERCC1-19007T>C, MTHFR-677C>T, MTHFR 1298A>C, DPYD IVS 14+1 G>A, single nucleotide polymorphisms in *GSTP1*, *XRCC1, ERCC1, MTHFR TSER* and *DPYD* genes were sequenced by pyrosequencing method. The polymorphism and progression-free survival times of each patient were subjected to Cox analysis to investigate the response to treatment after FOLFOX treatment and the effect of the relevant polymorphism.

Results: The progression-free survival times evaluated in the wild type (wt) and heterozygous (het) subgroups were found to be statistically significant in the XRCC1 G28152A (log-rank p=0.0113), and ERCC1 C8062A (log-rank p<0.000.1) polymorphisms.

Keywords: Colorectal cancer, SNP, FOLFOX, polymorphism, PFS

Introduction

Colorectal cancer is a common and deadly cancer worldwide. It is the third leading cause of cancer-related deaths (1). Almost one-third of patients have locoregional lymph node metastases at the time of diagnosis and are diagnosed at stage III. When treated with surgery alone, 40-50% of patients with locoregional disease develop recurrence or distant metastases due to micrometastases (2,3). Adjuvant chemotherapy aims to eliminate these micrometastases and ensure cure. For the first time in the literature, Moertel et al. (4) showed that 12 months of 5-fluorouracil (5-FU) and levamisole combination for stage III lymph node-positive colon cancer led to a 33% reduction in mortality rates. Following the demonstration of the survival advantage of the combination of 5-FU and leucovorin (LV) for 6 months, adjuvant chemotherapy based on 5-FU/LV for 6 months has become the standard treatment for stage III colon cancer (5). The Multicenter International Study of Oxaliplatin/5-FU/LV in Adjuvant Treatment of Colon Cancer trial found that adding oxaliplatin to 5-FU/LV resulted in a 7.5% increase in 5-year disease-free survival (DFS) and a 4.2% increase in 6-year overall survival, and recommended oxaliplatin-based chemotherapy as standard postoperative treatment for these patients (6). Gene polymorphisms and mutations reduce the efficiency of chemotherapy regimens by affecting the metabolism of drugs. Specific polymorphisms enhance the efficacy of some drugs, while others greatly increase the toxicity of these drugs. Therefore, they are thought to be effective in predicting clinical response to chemotherapy or chemotherapy-related toxicity in many cancer types. 5-FU acts via fluorodeoxyuridine monophosphate (FdUMP), a molecule that inhibits thymidylate synthase (TS). It increases the risk of various types of cancer by acting on functional polymorphisms in the *TS* gene. It enables nucleotide synthesis by altering the activity of the enzyme. Dual (2R) and triple (3R) repeating polymorphisms in the thymidylate synthase promoter end region (TSER) are described. *In vitro* and *in vivo* studies have shown that TS expression is overexpressed by the 3R allele of the TSER genotype. Thus, several epidemiologic studies on colorectal cancer risk and TSER 2R/3R polymorphism affecting Folate metabolism have been conducted but the results are inconsistent (7). Reduced 5-FU clearance and increased 5-FU toxicity have been observed in some colorectal cancer patients with *DPD* gene containing IVS14+1G>A polymorphism. When methylenetetrahydrofolate reductase (MTHFR) enzyme activity decreases, intracellular 5,10-methylenetetrahydrofolate (5,10-MTHF) levels increase and FU cytotoxicity may be increased due to this increase (8). Two types of single nucleotide polymorphisms (SNPs) frequently develop on this enzyme: C677T (rs1801133) and A1298C (rs1801131) (9). Alanine and valine change their positions as a result of the C677T transition, thus forming a more thermally unstable protein that reduces enzyme activity (10). The A1298C variant (Glu429Ala) can also reduce MTHFR enzymatic activity by causing a missense mutation. In C677T homozygous (TT) or heterozygous (CT) genotypic conditions, MTHFR enzyme activity is reduced and blood homocysteine levels are increased. When the A1298C enzyme is converted from the normal (AA) to the homozygous (CC) or heterozygous (AC) form, a decrease in MTHFR activity is observed, but no thermo labile protein is formed. Reduced enzyme activity results in high levels of 5,10-MTHF and thymidine, leading to increased DNA synthesis and repair. Therefore, MTHFR polymorphisms are thought to be protective against tumorigenesis (11,12). Oxaliplatin, a diaminocyclohexane derivative of cisplatin with higher water solubility and fewer toxic side effects, is actively used in colon cancer combination chemotherapy regimens (13,14). Oxaliplatin inhibits DNA replication and transcription by making covalent bonds to DNA. Oxaliplatin's mechanism of action and gene polymorphisms, families, Excision Repair Cross-Complementing group 1 (ERCC1), X-Ray Repair Cross-Complementing group 1 (XRCC1), glutathione Stransferases P1 (GSTP1) SNPs.

Methods

Patient Group

Patients with pathologically diagnosed colorectal cancer, stage IV according to WHO classification, who were admitted to Pamukkale University Faculty of Medicine, Department of Medical Oncology between 2014-2018 were included.

Genetic Evaluation

i. DNA Isolation

Blood samples of the patients included in the study group were taken 2-3 mL into hemogram blood tubes. DNA isolations were performed on Qiagen EZ1 advanced XL device using 200 µL of patient blood.

ii. Mutation Analysis

Polymorphism analysis of the patients' DNA was performed simultaneously with amplicon generation using Corbet 5Plex RT-PCR to monitor the number of amplicons. PCR products were then analyzed for polymorphism by pyrosequencing method using Qiagen Pyromark Q24 device. The SNPs included in the study were GSTP1-313A>G, XRCC1-G28152A, ERCC1- 8092C>A, ERCC1-19007T>C, MTHFR- 677C>T, MTHFR 1298A>C, DPYD IVS 14+1 G>A.

In this retrospective study, national and international ethical rules were complied with. This study approved Pamukkale University Ethic Committee with number of E-60116787-020-443991.

Statistical Analysis

COX analysis was used to investigate the effect of genetic mutations on patients' responses to treatment. Statistical analysis was performed with SPSS-21 and Graphpad programs. Results were evaluated at 95% confidence interval. $P<0.05$ was considered statistically significant.

Results and Discussion

We studied 38 patients, 27 men (71%) and 11 women (28%), with advanced and metastatic stage (stage IV) colorectal cancer who received 5-FU-LV-oxaliplatin (FOLFOX) chemotherapy regimens. In our study, the mean age of the patients was 68.94±8.83 years and the median age was 68.5 years. In this context, 71.05% of the study group consisted of male patients and in this context, the study group is consistent with the literature in terms of both age and gender distribution (15). Demographic and genetic characteristics of the patients are given in Table 1-3.

According to GSTP1 A313G, XRCC1 G28152A, ERCC1 C8062A, ERCC1 T19007C, MTHFR C677T, MTHFR A1298C, DPYD IVS 14+1 G>A, TSER polymorphism; in progression-free survival analyses performed in mutated (mut), wild-type (wt) and heterozygous (het) subgroups, statistically significant differences were found for XRCC1 G28152A, ERCC1 C8062A polymorphisms.

According to XRCC1 G28152A polymorphism, a statistically significant difference was found in the survival analysis between patients with G/G genotype and patients with G/A and A/A genotypes. The median DFS time was calculated as 18.09 months for patients with G/G genotype, 12.11 and 31.7 months for patients with G/A and A/A genotypes (log-rank p=0.0113) (Figure 1). When we consulted the literature, Huang et al.'s (16) studies on patients with metastatic colorectal cancer who received FOLFOX treatment showed a correlation between progression-free survival times in patients with wild type, heterozygous and mutant genotypes.

According to ERCC1 C8062A polymorphism, a statistically significant difference was found in the survival analysis between patients with C/C genotype and patients with C/A and A/A genotypes. The mean DFS time was calculated as 18.45 months for patients with C/C genotype and 21.54 and 3.42 months for patients with C/A and A/A genotypes (Log Rank p<0.000,1) (Figure 1). There are many studies on ERCC1 C8062A polymorphism and non-small cell lung cancer, colorectal cancer, nasopharyngeal cancer on both survival and progression-free survival. The data obtained in our study support each other with the literature data (17-19). This concordance suggests that the ERCC1 C8062A polymorphism is an important predictor of survival on treatment.

Although GSTP1 A313G, ERCC1 T19007C, MTHFR C677T, MTHFR A1298C polymorphisms did not show statistical significance in COX analysis for progression-free survival, it is clear that patients with wild-type genotype showed a later progression compared to mutant and heterozygous patients in accordance with the literature (Figure 1) (20,21).

This study suggests that treatment options for patients with advanced colorectal cancer should be tailored according to GSTP1 A313G, XRCC1 G28152A, ERCC1 C8062A, ERCC1 T19007C, MTHFR C677T, MTHFR A1298C, DPYD IVS 14+1 G>A, TSER polymorphisms. Patients with the mutant variant are recommended to prefer therapies other than FOLFOX for better survival. The limitation of this study is the patient population. This study could be strengthened with a larger number of participants.

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Table 1. Demographic characteristics of the patients

Characters	Patient (n=38) (%)
Age	
≤62	12 (33.3%)
>62	26 (66.6%)
Sex	
Male	27 (71.05%)
Female	11 (28.95%)
Smoking habit	
Smoker	34 (89.47%)
Non-smoker	4 (10.53%)

Table 2. Gene polymorphism analysis results of the patients (oxaliplatin)

Single nucleotide polymorphism	Metastatic, (n=38)
GSTP1 A313G	
Wild type	18
Heterozygous	14
Mutant	6
XRCC1 G28152A	
Wild type	22
Heterozygous	9
Mutant	7
ERCC1 C8062A	
Wild type	19
Heterozygous	10
Mutant	9
ERCC1 T19007C	
Wild type	14
Heterozygous	11
Mutant	13

Table 3. Gene polymorphism analysis results of the patients (5-FU)

Single nucleotide polymorphism	Metastatic, (n=38)
MTHFR C677T	
Wild type	18
Heterozygous	12
Mutant	8
MTHFR A1298C	
Wild type	18
Heterozygous	8
Mutant	12
DPYD IVS 14+1 G>A	
Wild Type	38
Heterozygous	-
Mutant	-
TSER	
Wild type	14
Heterozygous	14
Mutant	10

5-FU: 5-Fluorourasil

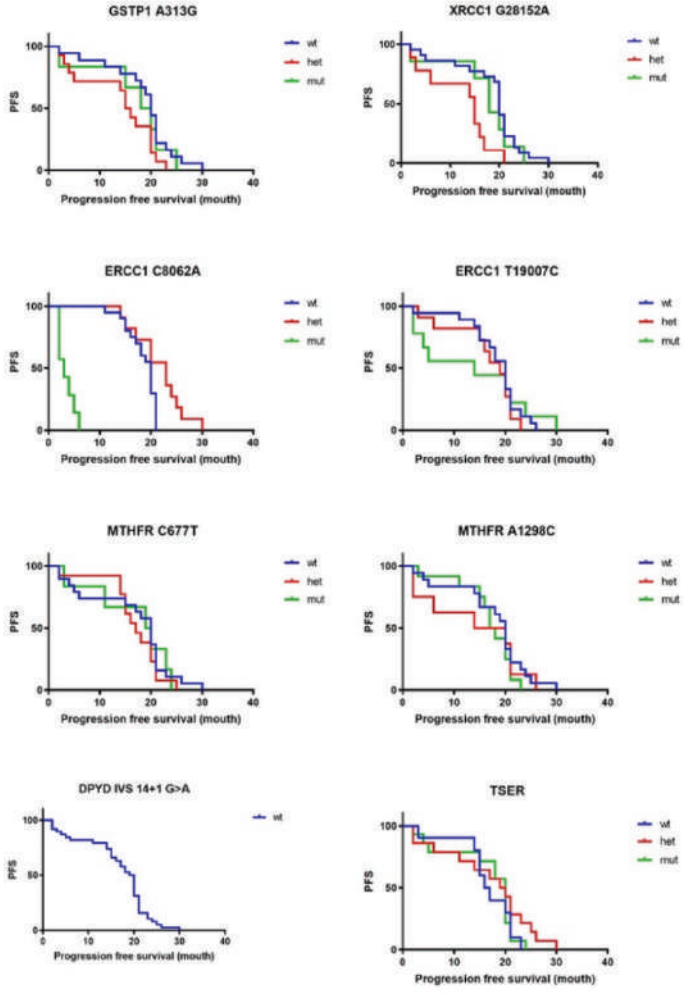

Figure 1. Progression-free survival curves according to GSTP1 A313G, XRCC1 G28152A, ERCC1 C8062A, ERCC1 T19007C, MTHFR C677T, MTHFR A1298C, DPYD IVS 14+1 G>A, TSER polymorphisms in advanced stage patients (wild type: wt, heterozygous: het and mutant: mut)

[OP-54]

Attention to Hypotonic Infant: Zellweger Family with Pathogenic Compound Heterozygous c.1804-2A>G and c.3693_3696delGTCA(p.Gln1231Hisfs)rs769836601 Variation in the *PEX1* Gene

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Introduction

Zellweger syndrome (ZS) is a rare autosomal recessive disease manifested by dysfunction of peroxisomes necessary for the oxidation of very long chain fatty acids (1). ZS, also known as cerebrohepatorenal syndrome, is a hereditary disease that affects many systems with serious neurological involvement. Hypotonia, seizures, jaundice, deterioration in liver function tests, malnutrition, and bone anomalies occur in the neonatal period. They have typical facial appearances; they have a high forehead, wide anterior fontanelle, epicanthus, upslanted palpebral fissure, and a hypoplastic supraorbital appearance. Retinopathy, cataracts, kidney cysts, adrenal insufficiency, and hearing impairment are other clinical features that may be observed. They usually die within the first year of life (2). Its incidence varies between 1/50,000 and 1/100,000. While the Canadian province of Quebec is the region with the highest incidence, Japan is the country with the lowest incidence. ZS has been associated with 13 different mutations in the *PEX* gene. *PEX1* and *PEX6* gene mutations were detected in 65% of the patients. *PEX* genes code for proteins called peroxins. Peroxins are necessary for the biogenesis of peroxisomes and ensure the transfer of proteins to the peroxisomal matrix and membrane. The absence of functional peroxisomes results in the absence of β-oxidation of very long-chain fatty acids and the accumulation of peroxisomal substrates. Tissue disorders occur in many organs (3). Next generation sequencing (NGS) is the process of multiplying, sequencing and accurately detecting the variations in large numbers of DNA fragments formed by cutting DNA through enzymatic reactions (4). In this study, we aimed to contribute to the literature by presenting the patient, who was diagnosed with ZS as a result of NGS, together with her family members.

Case Report

A 10-day-old baby was applied because of not being able to feed, bruising while feeding, and little movement. He was born at an external center at 38 weeks, G2P1Y1, weighing 3240 g. He cried as soon as he was born and was given to his mother. His mother stated that the baby had not been able to breastfeed well since birth, was crying slowly and was moving little. In his family history, it was learned that the first baby of the family died at 20 weeks of intrauterine period, and that the mother and father were their maternal uncle's children. In the physical examination of the patient, weight: 2700 g, height: 49 cm and head circumference: 34 cm were measured. Dysmorphic facial findings (protruding forehead, wide anterior fontanelle, flattened nasal root, epicanthus, upslanted palpebral fissure, small chin) were detected in the patient, whose vital signs were normal. Her respiratory system was normal, her respiratory rate was 50/min, and she had no gasping or groaning respirations. Circulatory system examinations were normal, heart rate was 130/minute, and no additional sounds or murmurs were heard. Externally, she appeared to be a girl, and the genitourinary system examination was normal. Abdominal examination was normal and no hepatomegaly or splenomegaly was detected. The patient was hypotonic on neurological examination and neonatal reflexes were absent. Blood hemogram, biochemical tests, CRP, thyroid function tests and kidney function tests were normal. From liver function tests: AST: 262 IU/L, ALT: 146 IU/L, GGT: 80 IU/L, PT: 28 sec, aPTT: 95 sec, INR: 2.3, T. dil: 16.3 mg/dL, D. bil: 6.8 mg/dL was detected. The patient was taken to the neonatal service with preliminary diagnoses of neonatal sepsis, metabolic disease, and biliary atresia. A blood culture was taken and the antibiotic combination ampicillin and gentamicin was started. There was no growth in blood culture. Although the patient was administered vitamin K, there was no decrease in aPTT value. Abdominal USG revealed no hepatomegaly or splenomegaly, but renal cortical cysts were detected. MR cholangiography was performed for the differential diagnosis of biliary atresia, and it was learned that the bile ducts were open. The factor 8, 9, 11 and the Von Willebrand factor levels were found to be normal. Brain MRI revealed pachygyria, perisylvian polymicrogyria, and corpus callosum agenesis. The chromosome analysis result of the peripheral blood sample taken from the patient was normal. Considering these findings, the patient's long-chain fatty acid level was sent with the preliminary diagnosis of ZS. It was determined that the C26 (hexacanoic acid) level increased, the C22 (docosanoic acid) level decreased, and the C26/C22 and C24/C22 ratio increased. As a result, to clarify the diagnosis, blood with EDTA was taken from the patient and the *PEX1* gene was scanned with NGS.

In the genetic examination, autosomal recessive (OR) class 2 pathogenic c.1804-2A>G mutations in the intronic region of the *PEX1* gene and pathogenic c.3693_3696delGTCA(p.Gln1231Hisfs)rs769836601 variation in the exon 23 of *PEX1* gene were detected and a diagnosis of ZS was made. Thereupon, segregation analysis was performed on the family and the pathogenic heterozygous c.3693_3696delGTCA(p.Gln1231Hisfs)rs769836601 variation in the father and the pathogenic novel c.1804-2A>G variation in mother were detected in the *PEX1* gene (Figure 1). The patient died in the 6th month of his life.

Methods

Physical examination, laboratory tests and imaging: The patient's physical and neurological examination were performed. Hemogram, biochemical tests, CRP, thyroid function tests, kidney function tests, liver function tests, long chain fatty acid level, factor 8, 9, 11 and Von Willebrand factor levels, and blood culture were performed from the blood. Abdominal USG, MR cholangiography, and brain MR imaging were performed.

Karyotype analysis: Heparinized peripheral blood samples were taken from the patients, and the metaphase plaques obtained after cell culture were banded with GTG banding and examined under a microscope for karyotype analysis.

STR fragment analysis: Maternal contamination was detected via STR fragment analysis performed on the mother using *D5S818, D7S820, FGA, D21S11, D8S1179, D13S317, VWA, D18S51, THO-1, D3S1358* STR markers.

Rapid aneuploidy test: Again rapid aneuploidy panel in the FISH (13, 18, 21, X ve Y) was performed using *LS1 13, LS1 18, LS1 21, CEPX VE CEPY* probs on CVS material.

Next-Generation Sequencing (NGS)

Genomic DNA was extracted from the peripheral blood samples and CVS. NGS was performed by capture of the coding regions and splice sites of target genes via Illumina custom enrichment panel. Annotation of detected variants was carried out using Illumina BaseSpace Variant Interpreter, InterVar, Franklin, VarSome, ClinVar, OMIM, and Pubmed. dbNSFP (contains SIFT, PolyPhen-2, LRT, Mutation Taster) were used to predict the pathogenicity of variants. Rare variants were classified according to the American College of Medical Genetics and Genomics criteria (5).

Discussion

ZS was named Cerebrohepatorenal syndrome due to the prominent findings in the cases when it was first described by De Lange and Janssen in 1949 (6). It was first reported by Hans Zellweger, after whom the disease is named, that it is caused by a mutation in the *PEX1* gene, which consists of 24 exons located at 7q21.2. Later, other peroxisomal diseases such as infantile refsum disease, neonatal adrenolipodystrophy, rhizomelic chondromalacia punctata were identified. However, since peroxisomes are found in all tissues except mature erythrocytes, it is known that they affect almost all tissues, starting from the intrauterine period. ZS is a rare disease and usually causes mortality within the first year (7). Whole exome sequencing is important to exactly understand the roles and functions of genes associated with different diseases in the etiopathogenesis (8-13).

In such diseases, it is very important to perform family segregation analysis studies and provide genetic counseling in order to have healthy children. In the genetic analysis performed on our patient's mother and father, c.1804-2A>G class 2 OR transitional heterozygous novel mutation in *PEX1* gene intron in the mother, c.3693_3696delGTCA(p.Gln1231Hisfs)rs769836601 class 2 heterozygous mutation in exon 23 of the *PEX1* gene in the father were detected. Two ways were suggested to the family to have a healthy child: The first way is to have a child through in vitro fertilization, the second way is to conduct genetic examination of the baby with CVS at the appropriate week after normal pregnancy is achieved. Termination if the baby is sick, and continuation of the pregnancy if healthy.

Since the mother and father were young, it was decided to perform CVS after pregnancy was achieved normally. No pathology was detected in the mother's cranium examination (Figure 2). No malignancy or intraepithelial lesion was detected in the cervical or vaginal cytology performed on the mother. When the mother became pregnant in 2019, CVS sampling was performed at the 12th week of pregnancy. Karyotype analysis from CVS material was normal (Figure 3). No maternal contamination was detected in the STR fragment analysis performed using *D5S818, D7S820, FGA, D21S11, D8S1179, D13S317, VWA, D18S51, THO-1, D3S1358* STR markers from the CVS material. Again, a normal hybridization pattern was observed in the FISH (13, 18, 21, X ve Y) in rapid aneuploidy panel performed on CVS material. After the genetic examination was normal, the pregnancy continued and they had a healthy daughter. In 2021, the mother became pregnant again and in the amniocentesis, compound heterozygous c.1804-2A>G and c.3693_3696delGTCA/p.Gln1231Hisfs mutations were detected in the fetus, no maternal contamination was detected, and upon the presence of prefrontal thickness in the fetus, a decision was made to terminate by discussing with the family.

Conclusion

NGS technologies have an crucial place in the detection of rare disorders and appear as an important technology for better understanding the etiopathogenesis of different hereditary diseases and making an accurate diagnosis. Genetic counseling is of great importance to develop an effective treatment strategy for hereditary disorders and to reduce the risk of birth with abnormalities so that couples can have a healthy child.

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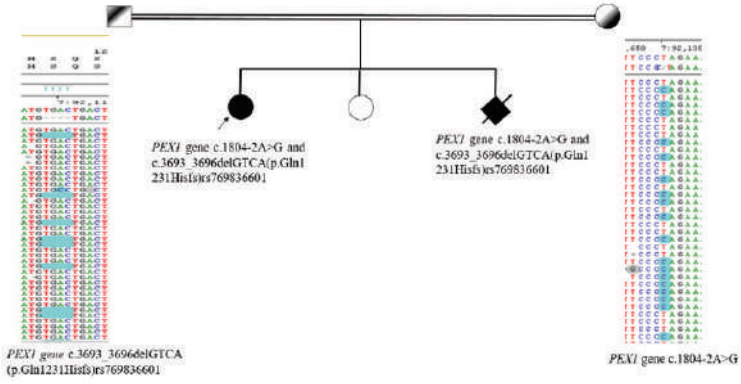

Figure 1. Family pedigree and detected variants

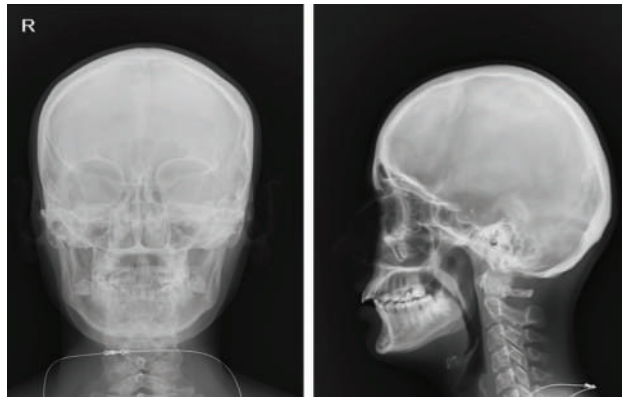

Figure 2. Image of mother's cranium

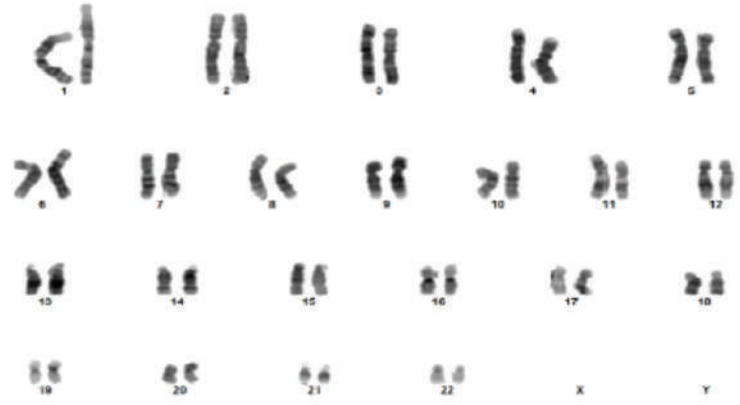

Figure 3. Karyotype analysis of fetal CVS material

[OP-55]

A New Translocation in a Case of Recurrent Pregnancy Loss: t(2;7)(q31;p21)

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Abstract

Recurrent pregnancy loss (RPL) is when a woman has three or more miscarriages in a row. Chromosomal abnormalities are frequently seen in RPL cases. In this case report, we detected a balanced t(2;7)(q31;p21) translocation in the male partner of a couple who applied to Erciyes University, Department of Medical Genetics with the indication of RPL. Balanced translocation is a type of chromosomal anomaly in which two chromosomes swap parts, but the total amount of genetic material remains the same. Balanced translocations are generally harmless to the carrier, but may increase the risk of RPL and other pregnancy complications. The female partner of the couple in this case had a normal karyotype (46,XX), but the male partner had a balanced translocation between chromosomes 2 and 7 (46,XY,t(2;7)(q31;p21)). For this reason, the couple's children were also examined in terms of family segregation. The t(2;7)(q31;p21) translocation is new to the literature and has not previously been reported as a cause of RPL. In our study, by performing Gene Ontology Enrichment Analysis, we determined that morbid genes in the relevant translocation regions are effective in embryo development. In the light of all the analyses, we conclude that the unbalanced transfer of the 46,XY,t(2;7)(q31;p21) karyotype to the next generation negatively affects the survival of the embryo due to the effect of morbid genes.

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8th INTERNATIONAL CONGRESS OF MEDICAL GENETICS

September 21-23th 2023

Poster Presentations

(P-01 — P-17)

[P-01]

A Rare *SLC6A1* Gene Variant in a Family with Intellectual Disability: A Case Report from Çanakkale, Türkiye

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Abstract

Introduction: SLC6A1-related neurodevelopmental disorder (SLC6A1-NDD) is autosomal dominant, featured by developmental delay, epilepsy, autism spectrum disorder, and attention-deficit/hyperactivity disorder. Language skills, particularly expressive language, are often more significantly affected than motor development. In this poster, we present the family's clinical findings and genetic analysis results with the SLC6A1 pathogenic variant.

Methods: Our patient, a 6-year-old girl, was referred to us because of trigonocephaly, speech retardation, autism spectrum disorder, and dysmorphic facial features. The patient's mother is 31 years old with epilepsy and intellectual disability. Her father is 41 years old with intellectual disability. There is no consanguinity between the patient's parents. We performed chromosome, microarray, and WES analysis for the proband and its family.

Results: Chromosome analysis was wild, no pathogenic or likely pathogenic variant that could explain the clinic was found in microarray analysis. Trio WES analysis detected a pathogenic heterozygous variant of SLC6A1 c.223G>A p.G75R in the proband and her mother.

Conclusion: The *SLC6A1* gene encodes a neuronal gamma-aminobutyric acid transporter protein that plays a crucial role in inhibitory neurotransmission in the brain. SLC6A1-NDD is a rare disease. Less than 500 have been reported worldwide. This case highlights the importance of comprehensive genetic analysis, such as WES, in identifying pathogenic variants associated with rare neurodevelopmental disorders.

[P-02]

A Delayed Diagnosis Case of a Patient with Treacher Collins syndrome

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Introduction

Treacher Collins syndrome [(TCS), OMIM #154500, ORPHA:861] is a genetic disorder affecting the craniofacial development. It is a rare mandibulofacial dysostosis characterized by a variety of facial abnormalities and is caused by mutations in the *TCOF1*, *POLR1D*, *POLR1C*, or *POLR1B* genes.

Case Report

Hereby, we are reporting a case of TCS with delayed diagnosis. A 16-year-old boy was consulted to our department by pediatrician due to dysmorphic features, including down-slanting palpebral fissures, eyelid coloboma, partial absence of lower eyelashes, bulbous nose, malar hypoplasia, dysmorphic ears, and oral cavity deformity. Additionally, he had bilateral hearing loss with normal intelligence. After preforming *TCOF1* gene sequence analysis, by next generation sequencing method, due to the patient's typical presentation, a likely pathogenic novel heterozygous frameshift variant [NM_000356.4: c.372dupA p.(Ala125fs)] was detected in the *TCOF1* gene.

Discussion

Despite clinical variability is common in TCS, this syndrome is usually diagnosed based on clinical examination and may be confirmed through molecular genetic testing. Our patient was an example of a delayed diagnosis, empathizing the importance of detailed physical examination and knowledge of dysmorphology in diagnosing rare diseases.

[P-03]

A Rare Case Report of Dysferlinopathy with Dominant Behaviour

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Abstract

A 39-year-old male patient presented with progressive muscle weakness. Gradual onset of fatigue started by age of six. Progressive difficulties in walking have been observed from the age of 10, and dysphagia toward solid foods manifested at 27 years of age. Subsequent to dysphagia bilateral upper extremity weakness transformed the patient's myopathic presentation into a global proximal extremity muscle weakness pattern. The dynamic and evolving nature of the patient's clinical presentation resulted in diverse diagnostic considerations over the course of follow-up, encompassing entities such as polymyositis, mitochondrial myopathy, and lipid storage myopathy. Notable magnetic resonance imaging findings were lipid atrophy, increased muscle signal, and enchondroma. EMG assessments indicated myogenic etiology and serum CK level was increasing. Biopsy showed increased lipid deposition. Clinical exome sequencing revealed a heterozygous pathogenic missense variant in DYSF:c.3172C>T(p.Arg1058Trp). Segregation analysis discloses the *de novo* occurrence of this mutation. *DYSF* gene dosage analysis with SNP-microarray was normal. Detected mutation is linked to the miyoshi muscular dystrophy type 1 (MMD1) and limb-girdle muscular dystrophy recessive type 2 (LGMDR2). It is noteworthy that dominant inheritance patterns within the realm of dysferlinopathy are exceedingly rare, with only a handful of documented cases globally. Our case represents a remarkable manifestation within this subset. Although recent research advocates against dichotomizing MMD1 and LGMDR2 into distinct cohorts for clinical assessment, the presence of dominant inheritance within one subgroup may potentially offer genetic differentiation between these two. Nevertheless, further in-depth investigations are requisite to establish the veracity of this phenomenon.

[P-04]

A Rare and Potentially Treatable Cause of Neurodegeneration: Cerebral Folate Deficiency

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Introduction

Cerebral folate deficiency is a progressive neurodegenerative disorder caused by biallelic pathogenic mutations in the *FOLR1* gene, which encodes the alpha subunit of the folate receptor in the choroid plexus. The disease manifests in late infancy with neurodevelopmental regression, hypomyelinating leukodystrophy, ataxia and epilepsy. Treatment with folinic acid has been reported to improve symptoms, and in some patients, even complete recovery has been reported.

Case Report

A 14-year-old male patient was referred to our clinic with epilepsy, neuromuscular regression and cerebellar atrophy. It was noted that he had no previous illness until the age of 7 years and his neurodevelopmental milestones were normal. He developed tremor and ataxia at the age of 7 and generalized tonic-clonic seizures at the age of 11. The seizures were partially controlled with dual antiepileptic treatment. At the age of 12, strabismus and walking difficulties were added to the patient's symptoms.

Electroencephalography revealed generalized periodic epileptic discharges, which were considered in favor of neurodegenerative processes. Cranial magnetic resonance imaging (MRI) showed hyperintense signal changes in the lentiform nuclei, cerebellar atrophy, and areas of no signal in the bilateral frontoparietal and basal ganglia on the SWI sequence, consistent with Wilson's disease (Figure 1). ATP7B sequence analysis performed for Wilson disease was normal. Then, clinical exome sequencing [SOPHiA Genetics Clinical Exome Solution v3 kit (CES; SOPHiA Genetics, Boston, MA) and sequenced on a NextSeq500 instrument (Illumina, San Diego, CA)] was performed for other possible etiology.

Clinical exome sequencing analysis revealed a homozygous non-sense variant, c.591C>A (p.Tyr197Ter), in the *FOLR1* (NM_016729.3) gene, which was classified as likely pathogenic (Figure 2). Following genetic diagnosis, low 5-methyltetrahydrofolate level in the cerebrospinal fluid supported the diagnosis. Oral folinic acid treatment was started.

Discussion

Cerebral folate deficiency due to FOLR1 mutations is a very rare disorder, with 33 patients described in the literature to date. This gene encodes the folate receptor-alpha (FOLRα), which is densely expressed in the choroid plexus and is responsible for the transport of 5-methyltetrahydrofolate across the blood-brain barrier. Loss of function variants in this gene disrupt folate transport to the brain and cause progressive neurodegeneration.

The homozygous variant of FOLR1 (NM_016729.3) c.591C>A (p.Tyr197Ter) has not been previously reported in the literature. Since this variant is located in the last exon of the protein, non-sense-mediated decay of the transcript is not clear. Although it has been suggested that this presumed truncated protein may be responsible for the late onset of symptoms in our patient, even in patients who have variant may cause a longer truncated protein is compatible with the typical course of the disease (1, 2). The late onset of symptoms in our patient may be related with diet, unknown modifier genes or obscure initial findings.

Cerebellar atrophy, found in 84% (28/33) of patients, is the most common MRI finding and may be important in the differential diagnosis. In addition, subcortical and periventricular white matter lesions, demyelination, hypomyelination, cerebral subcortical calcifications, basal ganglia calcifications, encephalomalacia, laminar necrosis, thin corpus callosum can be considered in the differential diagnosis of many diseases (3).

Conclusion

Early treatment with folinic acid has been shown to stabilize and even reverse neurodegenerative processes in some patients (3). For this reason, early diagnosis and treatment of this rare disease are of the utmost importance.

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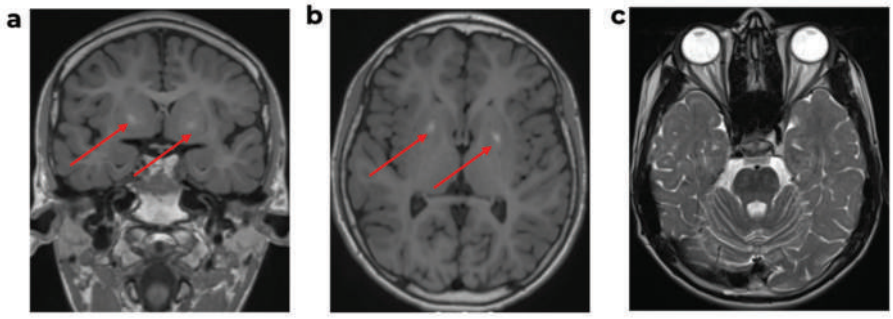

Figure 1. Cranial MRI results of the patient (a), (b) hyperintense areas in the lentiform nuclei on T1-weighted imaging (c) Prominence of cerebellar foliation

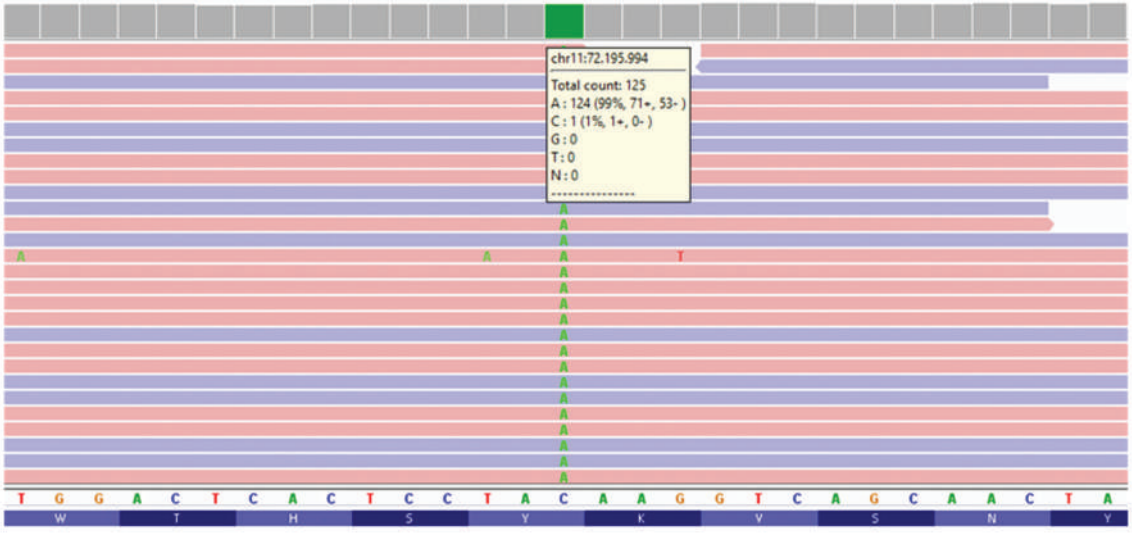

Figure 2. Integrative genomics viewer visualization of the homozygous FOLR1 (NM_016729.3) c.591C>A (p.Tyr197Ter) variant

[P-05]

A Rare Syndrome from Physical Examination to Diagnosis: Oculodentodigital Dysplasia

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Introduction

Oculodentodigital dysplasia (ODDD) is a rare disease caused by mutations in the *gap junction protein alpha-1 (GJA1)* gene located in the q22 region of chromosome 6, with high penetrance and variable expressivity (1). The *GJA1* gene, which has a coding exon, encodes the connexin 43 protein, weighing 43 kd and containing 382 amino acids. Although ODDD is mostly inherited in autosomal dominant (OD), autosomal recessive (OR) inheritance has also been described in recent clinical reports (2).

ODDD is characterized by symptoms such as craniofacial, neurological and ocular anomalies, type III syndactyly of the hands, phalanx abnormalities, diffuse skeletal dysplasia, enamel dysplasia and hypotrichosis (3).

Case Report

A 16-year-old male patient was referred to us from the department of dermatology due to keratoderma on both hands and soles of the feet, syndactyly and a dysmorphic appearance on the face. The patient had learning disabilities and early tooth loss. In the family history of the patient

who has a history of surgery due to undescended testicle; it was learned that his mother, his uncle and his uncle's son, and his maternal grandfather had a history of F4-5 (phalax) syndactyly in bilateral hands. On physical examination, high nasal root, retrognathia, blepherophymosis, short palpebral fissure, ala nasi hypoplasia, prominent columella, sawtooth appearance, F4-5 brachydactyly in bilateral hands, F4-5 cutaneous syndactyly in bilateral hands (operated), F4-5 camptodactyly in bilateral hands, F3-5 ulnar deviation in bilateral hands, bilateral plantar hyperkeratosis, and bilateral 2-3 partial cutaneous syndactyly in the feet were observed.

When the patient was consulted to the relevant departments for findings that may accompany the syndromic appearance, a slight increase in left ventricular wall thickness and left ventricular concentric hypertrophy (mild, not causing stenosis) were detected on ECHO.

At the same time, he was examined at an external center with a complaint of headache. In his brain CT examination, calcifications were randomly observed in both putamen.

When the patient's physical examination findings were scanned from databases, ODDD, oculodentodigital dysplasia (OR) and type 3 syndactyly were considered among the preliminary diagnoses. All preliminary diagnoses were due to GJA1 mutations.

OD inheritance was considered due to the vertical inheritance pattern in the family tree. With these preliminary diagnoses, DNA sequence analysis was performed on the patient with primers designed for the *GJA1* gene.

Discussion

GJA1 gene mutations can cause 6-allelic diseases inherited as OD and OR. With this case example, it can be seen that a pinpoint diagnosis can be reached in rare single-gene diseases by detailed anamnesis, system interrogation, determination of the inheritance pattern from the family tree, and physical examination findings in databases. Genotype-phenotype correlation can be made in allelic diseases with clinical evaluation.

Conclusion

A c.413G>A(p.Gly138Asp) heterozygous pathogenic variant in the *GJA1* (NM_000165.5) gene was detected in the patient.

Keywords: Oculodentodigital dysplasia, GJA1, type 3 syndactyly

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[P-06]

Diagnosis of Mandibuloacral Dysplasia in Clinical Heterogeneity of Scleroderma with Restrictive Dermopathy

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Abstract

Mandibuloacral dysplasia (MAD) is one of rare autosomal recessive syndromes characterized by postnatal onset growth retardation, craniofacial anomalies with mandibular hypoplasia, cutaneous pigmentation, lipodystrophy, rapid aging, and bone abnormalities with progressive osteolysis in the distal phalanges and clavicles. Typical feature of disease is that cases are completely normal at birth but symptoms develop as age progresses. Subtypes of MAD are defined as type A (MADA, OMIM#248370) and type B (MADB, OMIM#608612) which develops from *LMNA* and *ZMPSTE24* mutations respectively. Homozygous or compound heterozygous *LMNA* mutations create cellular stress through chromatin dynamics, and may represent a single disorder of varying degrees of severity. Detailed characteristics of MADA syndrome have not yet been clearly determined due to its rarity and limited number of announced cases. In this report, we present a case of 26-year-old female with unique MADA phenotype which shows high clinical heterogeneity. Whole exome sequencing (WES) analysis revealed a homozygous c.1580G>A (p.Arg527His) mutation in exon9 of *LMNA*. Additionally, a homozygous c.581T>C (p.Val194Ala) mutation was detected in exon6 of *SERPINB8*. *LMNA* mutation is associated with 11 different phenotypes in the literature and shows a high degree of clinical heterogeneity. For this reason, we diagnosed MADA with differential diagnosis method using genotype-phenotype data of human gene mutation database (HGMD). To evaluate phenotypic variability in the case, we evaluated our observations with MADA cases in the literature. This report aims to increase the number of reported MADA cases and strengthen genotype-phenotype correlations in cases.

Introduction

Mandibuloacral dysplasia (MAD) is a segmental form of progeria (1). MAD is a rare autosomal recessive syndrome characterized by dysmorphic findings, craniofacial anomalies, skeletal malformations, growth retardation, cutaneous pigmentation and metabolic disorders. Cases are completely normal at birth but growth retardation and dysmorphism develop as age progresses (2). Clinical signs of MADA(OMIM#248370) appear in early childhood while MADB(OMIM#608612) is defined by a more severe phenotype (3). MADA frequently arises from mutations in exons 8-10 (4). Laminopathy phenotypes of variants are quite heterogeneous. Currently, 3 autosomal recessive and 7 autosomal dominant primary laminopathies have been reported. In our clinic, we diagnosed a case with features of MAD. We performed WES analysis to determine genetic cause of clinical phenotype.

Case Report

Case was referred by dermatology with suspicion of progerioid syndrome. Complaints started in fingers at age of 2.5. growth retardation, mobility difficulties and skin atrophies became evident at age of 6. She was diagnosed with scleroderma and her complaints increased. There was a consanguineous marriage. Prenatal and postnatal were normal. Body height was 155 cm, head circumference was 52 cm and neuromotor abilities were normal. Sclerodactyly of fingers, dystrophy of fingernails and toenails were observed. There was bilateral pes planus, calluses on plantar surface, skin atrophy, superficial veins, calcinosis cutis in lumbar region and limited joint movements. It was remarkable that there was an excessive fat tissue in abdomen and submandibular region contrary to extremities. Mitral anterior leaflet prolapsed into left atrium in systole. Also, there were minimal tricuspid regurgitation, restrictive breathing, minimal thickening in pericardium, increased nodular density in fatty tissue thymus chamber in anterior mediastinum, mild mitral and aortic regurgitation (Figure 1). We determined early diagnosis as Scleroderma-Werner syndrome and/or an autosomal recessive syndrome.

Discussion

Mutations were detected in *LMNA* and *SERPINB8*. *LMNA* mutation was distinctively associated with mandibuloacral dysplasia in HGMD. *SERPINB8* mutations were associated with peeling skin syndrome 5(OMIM#617115) in OMIM.

LMNA mutations cause anomalies in nuclear structures (5). 8% of restrictive dermopathy cases are caused by mutation in *LMNA* (6). Similar to our case, clinical findings were reported as low birth weight, fragile-tight skin, joint contractures, pointed nose, micrognathia superficial erosions and vascular structuring. There were also secondary prominent superficial veins in atrophic skin of extremities and calcinosis cutis in lumbar region (1). Same mutation was reported in two children (7). MADA phenotype was observed tenuously in infancy, increased over time, and ocular proptosis developed in the boy. These are compatible with the initial period and characteristics of complaints in our case.

LMNA c.1579C>T mutation were reported with subtotal alopecia, absence of clavicle and ribs due to severe osteolysis and muscle damage (8). There was no muscle damage while alopecia and osteolysis were observed limitedly in our case. Therefore our case is a good example of allelic and clinic heterogeneity. An interesting MADA case that resemble limb-girdle myopathy were described in the literature (9). Clavicular hypoplasia and metabolic imbalance were not observed. Clinical features were reported as hypoplastic mandible, acroosteolysis, pointed nose, partial loss of subcutaneous fat tissue and progeric appearance. Case was evaluated as atypical laminopathy phenotype due to normal metabolic profile associated with general hypotonia.

SERPINB8 variant is also likely to affect phenotype. Family members, who had 2 different exons mutations, were examined with exfoliative ichthyosis (10). Hyperkeratotic plaques on palmar skin, superficial flaking on forearms and lower extremities without erythema and superficial peeling on dorsal skin of hands and feet were observed. There were calluses on plantar surface and calcinosis cutis in lumbar region in our case. However, the missense mutation in exon6 of our case has not been reported yet.

Conclusion

MADA necessitates a multidisciplinary basis. Mandibular hypoplasia, clavicular resorption, acral osteolysis, alopecia and lipodystrophy should be investigated carefully in clinic. We suggest comprehensive evaluation of progeroid syndrome outlook in pediatric patients with scleroderma-like disease. It would be possible to take measures to prevent osteolysis with early diagnosis.

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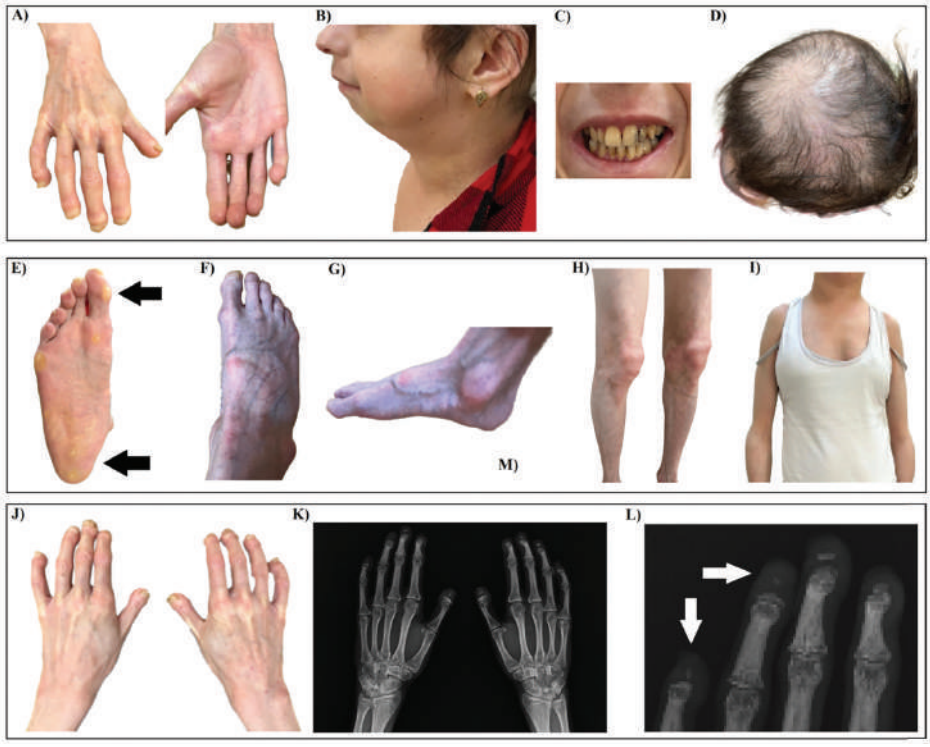

Figure 1. (A) Skin atrophy and hypoplasia of fingers, (B) mandibular hypoplasia, pinched nose and retromicrognathia, (C) crooked teeth, (D) subtotal alopecia, (E) plantar callus, (F) prominent superficial veins with dermal atrophy, (G) bilateral pes planus, (H) lipodystrophy, (I) sloping shoulders, (J) dystrophy of short distal phalanges and nails, (K) osteolysis of distal phalanges, (L) acroosteolysis

[P-07]

Legius Syndrome with a Preliminary Diagnosis of NF1-Like Syndrome: Case Report

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Introduction

Neurofibromatosis 1 (NF1) or Von Recklinghausen disease is a common autosomal dominant disorder that occures in 1 in 3,000 live births. Multiple caf´e-au-lait spots, freckling in skin fold areas, Lisch nodules, optic glioma and neurofibromas are the most common clinical findings of NF1. Other findings are short stature, macrocephaly, scoliosis, specific bone abnormalities, and increased risk of certain tumors. NF1 syndrome occurs as a result of an inactivating mutation in the *NF1* gene, located on chromosome 17q11.2 and encoding the neurofibromin protein. Neurofibromin protein is one of the main regulators affecting the complex RAS-MAP kinase signaling pathway. İnactivating mutations in the *NF1* gene results in activation of RAS-MAPK pathway, and leads to uncontrolled proliferation and an increase in the risk of malignancy. Later, many genes other than the *NF1* gene, which are associated with the RAS-MAPK signaling pathway, were found. Many of these genes cause syndromes called RASopathies, which have distinctive features as well as overlapping clinical findings. In 2007, a new RASopathy with a mild NF1-like clinic was discovered. In this new syndrome, a loss-of-function variant was detected in the *SPRED1* (OMIM# 609291) gene, which is a negative regulator of the RAS-MAPK signaling pathway. Firstly this syndrome was called NF1-like syndrome and it is now called Legius syndrome (OMIM # 611431). Findings are typically multiple

caf´e-au-lait spots and axillary freckling. Other findings include attention deficit, learning problems, and macrocephaly. Besides phenotypes of the patients are milder compared to NF1, the tumors typically observed in NF1 do not occur in these patients.

Case Report

A 29-year-old female patient, who was referred to us from the dermatology department with a preliminary diagnosis of NF, had many polymorphic brown skin spots on her body. The patient, who visited to the dermatology clinic with the complaint of hyperpigmented plaque on the lateral edge of the tongue, underwent a detailed physical examination in our clinic.

The patient, who had many caf'e-au-lait spots and axillary freckles on her body, did not have any tumors. whose eye examination of patient was evaluated as normal, the same skin findings were found in his mother, aunt and grandmother. There was no history of tumor in these people. A NF panel was taken from the patient and in this panel, there were *NF1, NF2, SPRED1* and *SMARCB1* genes.

Results

Genes within the panel were evaluated. As a result of NGS analysis, heterozygous c.C70T pathogenic variant was detected in the patient's *SPRED1* gene. This variant has also been evaluated as pathogenic by prediction programs such as Varsome and Franklin and is among the variants previously reported in the ClinVar database. After evaluation of the pathogenic variant obtained and the patient's clinic, our patient was diagnosed with Legius syndrome (OMIM# 611431). Genetic counseling was given to the patient in our genetics outpatient clinic, and it was recommended that the same variant be screened in people with similar clinical symptoms in the family.

Discussion

Legius syndrome is an autosomal dominant syndrome and occurs as a result of inactivating germline mutations in the *SPRED1* gene. In this syndrome, which can be seen in inherited or sporadic form, cafe' au-lait spots are typically observed, similar to the NF clinic, while there is no increase in the risk of developing tumors such as neurofibroma or optic glioma. The *SPRED1* gene, consisting of 8 exons, is located on the long arm of chromosome 15 and encodes the SPRED1 protein from the SPROUTY/SPRED family. SPRED1 protein plays a role as a negative regulator in the RAS-MAPK signaling pathway. In other words, loss-of-function variants in the *SPRED1* gene cause overactivation of the RAS-MAPK pathway.

In patients with a preliminary diagnosis of NF1, a detailed physical examination should be performed for differential diagnosis with NF-like syndromes. In addition, in patients with a preliminary diagnosis of NF1 and in whom no pathogenic or possible pathogenic variant can be detected in the *NF1* gene, NF-like syndromes should be considered and other genes affecting the RAS-MAPK pathway should be examined.

[P-08]

The Importance of Genetic Analysis in the Diagnosis of Complex Clinical Manifestations: A Case Report of Metabolic Rare Disease

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Abstract

Introduction: Hereditary coproporphyria (HCP) is a metabolic rare disease caused by heterozygous variants in the *CPOX* gene, with a prevalence of 1-9/1,000,000. A 54-year-old male patient was referred to our clinic due to dermatological, gastrointestinal, and cardiovascular symptoms triggered by certain medications and foods. The patient reported experiencing dermatological symptoms after using lansoprazole and rabeprazole, diarrhea following the consumption of acetylsalicylic acid, piracetam, and ginkgo biloba, and tachycardia after using pancreatic enzymes (pancreatin), N-acetylcysteine, and ursodeoxycholic acid. Additionally, he exhibited dermatological symptoms after consuming mushrooms, pepper, and peanuts. The patient's medical history includes hypothyroidism, vertigo, and a history of non-invasive papillary urothelial bladder tumor. In this study, we aim to present a patient with a detected pathogenic variant in the *CPOX* gene.

Methods: Clinical exome sequencing was performed performed from blood sample, and a heterozygous missense variant, c.1339C>T;p.R447C (NM_000097.7), was identified in the *CPOX* gene.

Discussion: HCP is a metabolic disorder characterized by gastrointestinal, cardiovascular, and neurological symptoms triggered by specific medications and foods. The patient's gastrointestinal, cardiovascular, and dermatological symptoms consistent with coproporphyria, as well as autoimmune and neurological manifestations like hypothyroidism and vertigo, may be associated with this genetic variant. However, it is possible that other genetic and environmental factors may contribute to these findings. This case emphasizes the significance of genetic testing in diagnosing rare and complex clinical manifestations.

[P-09]

A Rare Case: Smith-Magenis Syndrome

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Abstract

Smith-Magenis syndrome (OMIM #182290) is a complex neurodevelopmental disorder, with an incidence of 1/15,000-1/25,000, involving multiple organs and systems and usually occurs sporadically. This syndrome is characterized by typical dysmorphic features, speech delay, mental retardation, behavioral problems, sleep disturbance, renal and cardiac defects in some patients. The majority of cases are caused by interstitial micro-deletions in the 17p11.2 chromosome region containing the *retinoic acid induced 1* gene. We present a 2.5-year-old girl who stands out with her dysmorphic findings and delay in neuromotor steps. In the physical examination of the patient, brachycephaly, prominent forehead, square shaped coarse facial appearance, uplifted ear lobes, upslanting palpebral fissures, characteristic downturned tented upper lip vermilion, and brachydactyly were detected. In microarray analysis, a 3.6 Mbp deletion was detected in the 17p11.2 chromosome region. This case has once again demonstrated the importance of physical examination and molecular genetic methods in the diagnosis of genetic diseases.

Keywords: Smith-Magenis syndrome, 17p11.2 deletion syndrome, *RAI1* gene, microarray

[P-10]

Hereditary Hyperekplexia: Three Patients from Kayseri, Middle Anatolia and Three Different Genetic Findings by Different Methodology

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Abstract

Hereditary hyperekplexia (HPX), a neuronal disorder caused by genetic defects leading to dysfunction of glycinergic inhibitory transmission, is mainly characterized by startle responses to unexpected sensory stimuli and stiffness. HPX, a rare and underdiagnosed disorder, is manifest after birth and commonly improves with age. Establishing the correct diagnosis early is essential so that proper management may be initiated to reduce the risk of complications, such as potentially life-threatening apnea during episodes of stiffness. Defects in GLRA1 are the most common cause of HPX, inherited both in an autosomal dominant and autosomal recessive manner. Sequence analysis (95%) is the main method for detection of pathogenic variants of probands. Also copy number variations (CNVs) (5%) plays role in etiology. We here report independent three Turkish patients with hyperekplexia which stems from GLRA1 related phenotypes and we confirm mostly known genetic background of HPX by different methods in our outpatient clinic. Whole exome sequencing-CNV, microarray analysis revealed that previously reported homozygous deletion of exons 1-7 of the *GLRA1* gene in patient 1. This genetic changes thought to be probably the founder mutation in Turkish-Kurdish populations. In patient 2, homozygous c.277C>T p.Arg93Trp variant in the *GLRA1* gene was found. In patient 3, microarray analysis revealed a 299 kb deletion at the q33.1 region of the chromosome 5 which is *GLRA1* gene located in. The fact that the results of three unrelated patients in one center can be considered in terms of planning the examination for deletion/duplication analyzes first in patients with GLRA1-related phenotypes in the Turkish population.

[P-11]

Case Report: Patient with Merosin-Deficient Congenital Muscular Dystrophy with Occipital Lissencephaly

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Abstract

Merosin-deficient congenital muscular dystrophy type 1A (MDC1A) (OMIM 607855) is an autosomal recessive disorder characterized by severe muscular dystrophy, which is typically associated with abnormal white matter. MDC1A is caused by loss of function mutations in the *LAMA2* gene located at chromosome 6q22. Laminin is a major component of the basement membrane. It is thought to mediate the attachment, migration,

and organization of cells into tissues during embryonic development. Here, we described one Turkish female patient who has intellectual disablity, contractures, muscle weakness and magnetic resonance imaging (MRI) result compaible with merosine deficient dystrophies. Molecular analysis of the patient's *LAMA2* gene revealed homozygous frameshift pathogenic mutation c.3630del p. (Ile1210Metfs*14) in exon 25. A 9-year-old female patient was consultated to our department with mentioned for evaluation. Brain MRI revealed diffuse symmetrical involvement of subcortical and deep white matter pathological signal change, diffuse thinning of the brain stem and bilateral occipital lissencephaly. The patient's clinical and radiographic features were compatible with MDC1A, which was confirmed by gene studies identifying the laminin gene mutation. It is expected to result in protein dysfunction. Loss-of-function variants in LAMA2 are known to be pathogenic. So the definitive diagnosis is estabilshed as MDC1A.

[P-12]

Duplication of 1q21.3q25.3 in a Newborn with Multiple Congenital Anomalies

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Introduction

Trisomy 1q is a rare chromosomal anomaly syndrome, resulting from the partial duplication of the long arm of chromosome 1, with a highly variable phenotype principally. The major semptoms include short stature, intellectual disability, craniofacial dysmorphism (macro/microcephaly, prominent forehead, posteriorly rotated, low-set ears, abnormal palpebral fissures, microphthalmia, broad, flat nasal bridge, high-arched palate, micro/retrognathia), cardiac defects and urogenital anomalies. Patients may also present cerebral (e.g. ventriculomegaly) and gastrointestinal malformations, as well as dystonic tremor and recurrent respiratory tract infections. There is a wide spectrum of clinical manifestations due to the great variability in the extent of the duplication size. In this presentation, we aimed to present a case with 1q21.3q25.3 duplication.

Case Report

Our genetic department was consulted by the pediatrics neonatal intensive care unit for a 7-day-old newborn who had respiratory difficulties, a syndromic facial appearance, and ambiguous genitalia. On clinical examination, length was 53 cm (90-97p), weight was 3290 g (50p) and head circumference was 36 cm (75-90p). The patient's physical examination revealed microphthalmia, blepharophimosis, telecanthus, epicanthus inversus, hypertelorism, broad, flat nasal bridge, micro/retrognathia, anteverted nares, brachycephaly, short-broad neck with redundant nuchal skin, glossoptosis, bilateral incomplete cleft lip, posteriorly rotated, low-set ears, macrotia, auricular skin tag in the left ear, bilateral clenched hands, overlapping fingers on left toes, talipes calcaneovalgus, ambiguous genitalia. Cranial MRI displayed the patient had a ventriculomegaly. Echocardiography showed left ventricular hypertrophy, minimal tricuspid regurgitation and secundum ASD.

Results

Karyotype analysis was performed from the patient's peripheral blood sample and was determined as 46,XX,dup(1)(q21q25). Microarray analysis showed duplication in the 1q21.3q25.3 region with a size of 28,76 Mb. The karyotype analysis from the parent's peripheral blood sample was performed for the explanation of this duplication. The mother's karyotype analysis was normal and the father's karyotype analysis showed 46,XY,inv(1)(q21q31).

Conclusion

In most cases, the duplication is the result of an unbalanced translocation with a possible imbalance of the other participating chromosome. It is difficult to evaluate the contribution of the 1q trisomy to the phenotype in cases involving another chromosome. Cases with pure partial trisomy 1q provide an opportunity to better define the partial trisomy 1q syndrome. The extent of the partial 1q trisomy syndrome, which is a rare condition, can be assessed using array-CGH methods. In trisomy 1q syndrome, no duplication involving just this area has ever been documented. This case will add to the body of knowledge regarding how to diagnose people with comparable phenotypic features or regional duplications.

Keywords: 1q21.3q25.3, 1q duplication, array-CGH

[P-13]

The Case with the Novel NALCN Variant

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Abstract

In this study, we present a case with a milder phenotype associated with a novel variant of the *NALCN* gene. A 5-month-old male patient was referred to our department by pediatric neurology due to hypotonia, developmental delay, and hypospadias. The patient, reported to have been born to a 30-year-old mother at 38+3 weeks of gestation, with a birth weight of 2950 grams and a length of 50 cm, had parents from a neighboring village, with no known consanguinity. The patient started holding his head at 8 months and began sitting without support at 14 months. The patient's body weight was -2.48 SDS, and head circumference was -1.99 SDS. Marked hypotonia, microcephaly, ptosis and amblyopia in the left eye, a widow's peak hairline, restricted right gaze, brachycephaly, prominent philtrum, overriding toes, and contractures in the hands and feet were observed. EEG and echocardiography did not reveal any abnormalities. The karyotype result from the patient was 46, XY, and Prader-Willi FISH tests yielded normal results. Subsequently, SMA-MLPA analysis was performed and resulted in 2 copies. A clinical exome panel was performed due to hypotonia, revealing a homozygous likely pathogenic novel variant c.1495dupA (p.Ile499AsnfsTer26) in the 13th exon of the *NALCN* gene. The variant in the patient was confirmed as homozygous by Sanger sequencing. The parents and the elder male sibling were found to be heterozygous carriers of the variant. Common clinical features observed in patients with biallelic NALCN mutations include severe hypotonia, intellectual disability, speech delay, epilepsy, and optic atrophy. In the literature, significant hypotonia is reported in the infantile period in most cases. However, our patient had head control at 8 months and was able to sit without support at 14 months. It was learned that supported steps were present after physical therapy during follow-up. Seizures are typically reported in patients after the age of 3-4 years. Our patient, who is currently 30 months old, has no history of seizures, which is presumed due to his age. Some cases require gastrostomy due to feeding problems, and our patient experienced feeding difficulties with solid foods, leading to cachexia. During the patient's follow-up, muscle atrophy and dystonia were observed. While atrophy was an expected finding, dystonia had not been previously reported. Therefore, a new phenotype is considered possible in this regard.

[P-14]

A Case of Cardiophaciocutaneous Syndrome without Cardiac Manifestations

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Abstract

Cardiophaciocutaneous syndrome [(CFC); OMIM: 615279] is a developmental disorder resulting from heterozygous mutations in the *MAP2K1* gene located on chromosome 15. It is characterized by distinctive craniofacial features, cardiac anomalies, hair and skin abnormalities, postnatal growth retardation, and hypotonia. Developmental and epileptic encephalopathy-36 (OMIM: 300884) is a neurodevelopmental disorder caused by heterozygous/hemizygous mutations in the *ALG13* gene on the X chromosome, resulting in severe delays in psychomotor development. A 6-year-old male patient was referred to our department from the neurology department with complaints of epilepsy, developmental delay, inability to speak, and walk. The patient, who had third-degree consanguineous parents, had no specific family history. Physical examination revealed strabismus, downward-slanting palpebral fissures, a broad nasal bridge, fragile hair, and relative macrocephaly. Clinical exome sequencing analysis of the patient detected a pathogenic missense heterozygous variant (c.389A>G, p.Tyr130Cys) in the *MAP2K1* gene, a missense hemizygous variant of unknown significance (c.49A>G, p.Ile17Val) in the *ALG13* gene, and a missense homozygous variant of unknown significance (c.650A>G, p.Glu217Gly) in the *PEX1* gene. Metabolic tests ruled out peroxisomal diseases in our patient. Considering the prominent features of neurodevelopmental delay and dysmorphic appearance, it is important to recognize the potential phenotypic contribution of the *ALG13* gene alongside the suspicion of CFC syndrome attributed to the *MAP2K1* gene. Notably, our patient did not exhibit any of the major cardiac anomalies typically seen in CFC syndrome, which makes this case noteworthy from that perspective.

[P-15]

Familial Hyperkalemic Periodic Paralysis: The Utility of History Taking and Pedigree Drawing in the Diagnostic Odyssey of a Treatable Disease

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Introduction

Hyperkalemic periodic paralysis (HPP) is a rare, autosomal dominant condition leading to episodes of muscle weakness or paralysis.

Case Report

A fourteen-year-old boy suffering from paroxysmal generalized muscle weakness was admitted. He was born to non-consanguineus parents and his past medical history was unremarkable. His family history revealed multiple effected relatives including his sister, father, uncle, cousin, grand mom and her siblings. The detailed questioning revealed that the episodes were triggered by hunger, excessive physical exercise and food intake such as melon, banana and grapes and resolved after drinking sugary water. During hospitalization, a thirty-minute attack was observed. The serum potassium level was found 5.7 mEq/L (normal: 3.5-5.0 mEq/L) during the attack, while his baseline serum potassium level was 4.1 mEq/L. The clinical history raised the pre-diagnosis of familial HPP which is an autosomal dominant disease compatible with his pedigree. Acetazolamide therapy was initiated and he had no further attacks after treatment. The single gene analysis of *SCN4A* gene revealed a heterozygous c.2111C>T mutation and confirmed the preliminary diagnosis. *SCN4A* gene mutation alters the function of sodium channels in skeletal muscle cells with an influx of sodium ions and increases the release of potassium from muscle cells causing an inability for skeletal muscles to contract and eventually developing into muscle weakness or paralysis.

Conclusion

Familial HPP is a rare but treatable autosomal dominant neurological condition. Careful history taking and pedigree drawing enables the establishment of a preliminary diagnosis and the selection of the appropriate genetic testing, thus shortening the diagnostic odyssey.

[P-16]

Kabuki Syndrome; Clinical and Genetic Diagnosis

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Introduction

Kabuki syndrome is characterized by long palpebral fissures, eversion of lateral third of lower eyelids, thick eyelashes, ptosis, broad, arched eyebrows, sparse eyebrows, depressed nasal tip, short nasal columella, large, cupped ears, cleft lip and/or palate. It is a rare, multisystemic, genetic disease characterized by a peculiar facial appearance, postnatal growth retardation, mild-moderate mental retardation, hypotonia, skeletal anomalies and the persistence of fetal fingertip pads. Heterozygous mutations in the *KMT2D* gene on chromosome 12q13 are the most common cause of KS and are inherited in an autosomal dominant manner. Mutations in the *KDM6A* gene are inherited X-linked. In this study, we presented 4 isolated cases with dysmorphic facial appearance, microcephaly and developmental delay.

Case Reports

Case 1: A 1-year-old girl presented with dysmorphic findings, hydrocephalus and elevated LFT. The patient, whose prenatal follow-up was unremarkable, was admitted to intensive care for 1 month due to respiratory distress and seizures. On examination, his weight was 6 kg (-3.51 SDS), height: 66 cm (-3.19 SDS), head circumference: 39 cm (-5.3 SDS). Characteristic facial dysmorphic findings such as microcephaly, sparse lateral eyebrows, long eyelids, flattened nose tip, and high palate were detected (Figure 1A). Her cardiovascular, respiratory, abdominal and neurological examinations were unremarkable. ECHO revealed peripheral pulmonary stenosis and minimal pleural effusion. In the genetic analysis of the patient who was considered to have Kabuki syndrome, c.4521C>A heterozygous mutation was detected in the 16th exon of the *KMT2D* gene.

Case 2: An 18-month-old female patient applied to us due to cleft palate-lip anomaly and dysmorphic appearance. On examination, weight: 5.1 kg (-5.6 SDS), height: 66 cm (-4.7 SDS), head circumference: 38 cm (-6.9 SDS), microcephaly, cleft lip and palate anomaly, long eyelids, long eyelashes, dysplastic, large protruding ears, fetal fingertip pads, brachydactyly, nail dystrophy and hypotonicity were present (Figure 1B). Echocardiography showed large VSD, pulmonary hypertension and hypoplastic right ventricle, and cranial magnetic resonance imaging (MRI) showed corpus callosum

hypoplasia and septum pellucidum agenesis. The patient, who had left renal agenesis on renal ultrasonography, left hip dislocation on Hip Ultrasound, hypsarrhythmia and mixed epileptic spasms on electroencephalography, was receiving antiepileptic treatment due to West syndrome and levothyroxine treatment due to central hypothyroidism. Karyotype analysis found 46,XX. No mutations were found in *KMT2D* and *KDM6A* gene Sanger sequence analysis.

Case 3: A 27-month-old female patient was referred to us due to dysmorphic appearance, developmental delay and hearing loss. The patient's prenatal history was unremarkable and her family history was unremarkable. On examination, the patient weighed: 12 kg (34 kg), height: 86 cm (16 cm), head circumference: 45 cm (-2.2 SDS). scoop-shaped, nasal root prominent, eyelids long, eyelashes long, microcephaly and hypodontia were observed (Figure 1C). There were bilateral gynecomastia, fetal pads on the fingertips, brachydactyly and nail dystrophy in the hands and feet. The patient's other system examinations were normal. Left hip subluxation and hemangioma on the left arm were detected. There was bilateral mild conductive hearing loss. Dural ectasia was detected in lumbar MRI. Chromosome analysis 46,XX. In CES analysis, the frameshift c.2091dup variant was detected as heterozygous in the *KMT2D* gene.

Case 4: A 13-year-old male patient applied to us with epilepsy, developmental delay, blue sclera and dysmorphic findings. He had a history of surgery for gastric hernia when he was 4 months old, undescended testicle at 6 months, and aortic coarctation when he was 20 months old. In the physical examination of our patient, his weight was 23.5 kg (-3.2 SDS), height: 119 cm (-5.2 SDS), head circumference: 48 cm (-4.99 SDS). Face is thin, long, bitemporal stenosis, eyebrows are sparse in the distal part, scleras are blue, eyes are large, eyelids are long, ptosis, nasal root is wide, palate is narrow and high, tooth alignment and shape are distorted, ear large folds are reduced, ear lobe is very large, kyphosis, mild pectus excavatum, fingertip fetal pads, brachydactyly, and nail dystrophy were detected (Figure 1D and 2). ECO revealed operated VSD, aortic coarctation, aortic insufficiency and mitral insufficiency. As a result of CES analysis, in the *KMT2D* gene; The intronic c.14515+5G>T variant was detected as heterozygous.

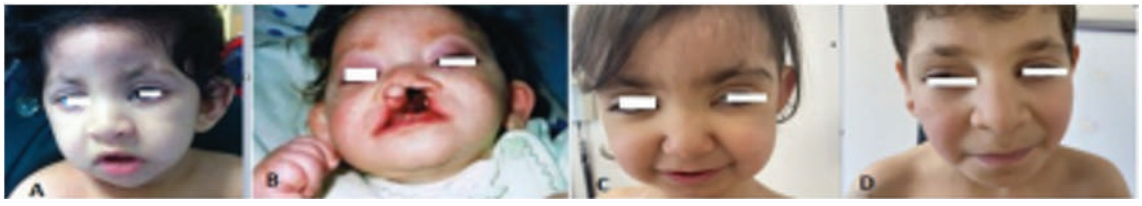

Figure 1. (A) Facial features of our 1-year-old patient, (B) 18-month-old patient with cleft palate-lip anomaly, (C) 27-month-old female patient, (D) 13-year-old patient with dysmorphic features on examination

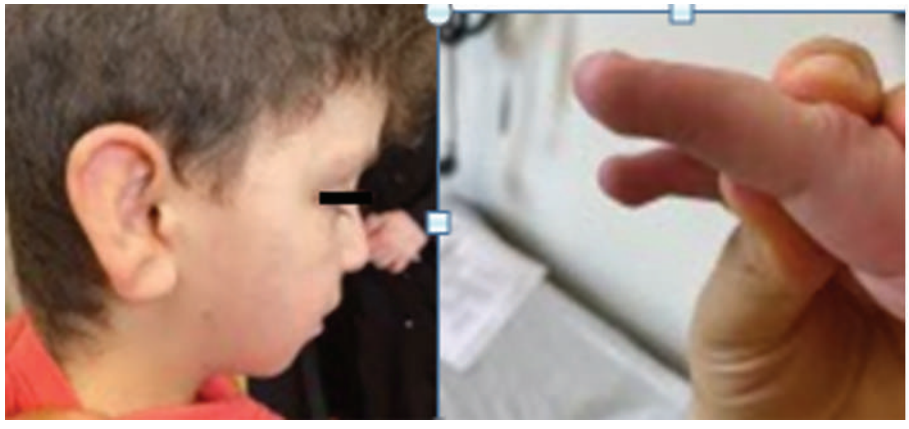

Figure 2. Presence of persistent fetal pads and large ears and ear lobe seen in our 4th case

[P-17]

A Rare Cause of Nephrolithiasis: Primary Hyperoxaluria Type 1

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Introduction

Primary hyperoxaluria is a rare autosomal recessive (OR) disease that occurs due to an increase in endogenous oxalate production. Its prevalence is 1 in a million. It occurs due to defects in glyoxalate metabolism. Three types have been described so far. Primary hyperoxaluria type 1, the most common type, occurs due to alanine glyoxalate aminotransferase (AGT) deficiency. This enzyme is encoded by the *AGXT* gene. The function of this enzyme is to catalyze the transamination of glyoxalate to glycine while converting pyruvate to alanine (1). When this enzyme does not function, the amount of endogenous glyoxalate increases. The amount of calcium oxalate excreted in urine also increases, causing clinical conditions ranging from recurrent urinary tract infections to nephrocalcinosis, nephrolithiasis, chronic renal failure and end-stage renal disease. When calcium oxalate load exceeds the urinary excretion capacity, it accumulates in various organ systems, causing systemic oxalosis (2). Diagnosis is made based on clinical history, urinary ultrasonography, urine oxalate amount and genetic examination. In necessary cases, it may be necessary to perform a liver biopsy and measure the AGT enzyme level (3). Whole exome sequencing has an important place in understanding the role of genes associated with different diseases in the etiopathogenesis of the disease (4-8). We would like to contribute to the literature by presenting a case about this very rare disease.

Case Report

A 10-year-old male patient was brought to the hospital with complaints of difficulty urinating and side pain for approximately 2 months. Abdominal pain is usually in the form of right-side pain. He stated that he had nausea, vomiting, enuresis, dribbling and intermittent urination. There was no dysuria, constipation or encopresis. He had no history of drug use and no known disease. He had a nephrolithotomy operation 1 year ago. In the family; it was learned that the grandfather, father, and cousins had a history of kidney stones, and the aunt had a history of chronic kidney failure. There was a 3rd degree consanguinity between the parents. The patient's FM scan revealed blood pressure: 100/50 mmHg, weight: 43.5 kg (74 p), height: 152.5 cm (91 p). Neurological examination, head and neck examination, circulatory system and respiratory system examination were normal. There was no rebound, guarding or tenderness on abdominal examination. Urinary system examination was normal. In the patient's examinations, creatinine: 0.56 mg/dL, Ca: 10.2 mg/dL, P: 4.46 mg/dL, ALP: 301 iu/L, PTH: 34.9 pg/mL, uric acid in spot urine/creatine ratio: 0.55; calcium/creatine: 0.44, 110 erythrocytes were detected in each area in the complete urinalysis. There was no growth in urine culture. In the urinary system USG, multiple calculi were observed in the right kidney, the largest of which was 12 mm in size in the pelvis, the calyces were dilated and the cortex was thinned in the upper pole. It was reported as follows: "The left kidney is normal and 12 mm of calculus was observed in the bladder lumen." In the stone analysis performed on the patient, it was learned that he had calcium oxalate stones. The patient's chromosome analysis was normal. Considering family history and stone analysis, *AGTX* gene was scanned with Next Generation Sequencing (NGS) with the preliminary diagnosis of primary hyperoxaluria. In NGS analysis, c.557C>T(p.Ala186Val) (rs117195882) and c.590G>A (p.R197Q) (rs346641134) compound heterozygous mutations were detected in exon 5 of the *AGXT* gene and the patient was diagnosed with primary hyperoxaluria type 1. The patient was started on ibuprofen and pyridoxine and was followed up.

Discussion

Primary hyperoxaluria type 1 is a hyperoxaluria condition that occurs as a result of the malfunction of the AGT enzyme due to the mutation in the *AGXT* gene. The clinical picture is wide ranging from recurrent urinary tract infection to end-stage renal failure. When the amount of oxalates exceeds the excretion capacity of the kidneys, it accumulates in other organs and oxalosis occurs. It most commonly affects bones, joints, skin, soft tissues, retina, heart, vessels, peripheral and central nervous systems. Oxalosis is a very important condition that affects mortality and morbidity (9). Symptoms usually begin around age 10. It was reported that 90% of the patients had nephrolithiasis and 48% had nephrocalcinosis at the time of diagnosis (10). Unfortunately, 30% of patients have been reported to have end-stage renal disease at the time of diagnosis. Definitive diagnosis is made by detecting the *AGXT* gene mutation and measuring the AGT enzyme level in liver biopsy. After diagnosis, patients' renal functions can be protected by high fluid intake, calcium oxalate crystallization inhibitor, and pyridoxine treatment. Liver and kidney transplantation is recommended together in patients who progress to end-stage renal failure (9). We diagnosed our patient with primary hyperoxaluria type 1 by detecting *AGXT* gene mutation at the age of 10. Our patient had nephrolithiasis at the time of diagnosis. We started treatment for nephrolithiasis and are following it with pyridoxine and ibuprofen treatment.

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