

Article

Not peer-reviewed version

Evaluation of Serum FGF21 Levels in Patients with Mitochondrial Aminoacyl-tRNA Synthetase Deficiency

[Sebnem Tekin Neijmann](#) , Dilek Gunes , Meryem Karaca , [Volkan Karaman](#) , [Mehmet Cihan Balci](#) , [Gulden Fatma Gokcay](#) , [Asuman Gedikbasi](#) *

Posted Date: 31 July 2025

doi: 10.20944/preprints202507.2586.v1

Keywords: mitochondrial disease; mitochondrial aminoacyl tRNA synthetase deficiency; FGF21; AARS2; EARS2; DARS2; SARS2; WARS2



Preprints.org is a free multidisciplinary platform providing preprint service that is dedicated to making early versions of research outputs permanently available and citable. Preprints posted at Preprints.org appear in Web of Science, Crossref, Google Scholar, Scilit, Europe PMC.

Copyright: This open access article is published under a Creative Commons CC BY 4.0 license, which permit the free download, distribution, and reuse, provided that the author and preprint are cited in any reuse.

Disclaimer/Publisher's Note: The statements, opinions, and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions, or products referred to in the content.

Article

Evaluation of Serum FGF21 Levels in Patients with Mitochondrial Aminoacyl-tRNA Synthetase Deficiency

Sebnem Tekin Neijmann ¹, Dilek Gunes ², Meryem Karaca ², Volkan Karaman ³, Mehmet Cihan Balci ², Gulden F. Gokcay ² and Asuman Gedikbasi ^{4,*}

¹ Department of Interdisciplinary Rare Diseases, Institute of Health Sciences, Istanbul University, Istanbul, Turkey; sebnemtekin@gmail.com

² Department of Pediatrics, Division of Pediatric Nutrition and Metabolism, Istanbul Faculty of Medicine, Istanbul University, Istanbul, Turkey; dilek.gunes@istanbul.edu.tr

³ Department of Medical Genetics, Istanbul Faculty of Medicine, Istanbul University, Istanbul, Turkey; volkan.karaman@istanbul.edu.tr

⁴ Department of Pediatric Basic Sciences, Institute of Child Health, Istanbul University, Istanbul, Turkey

* Correspondence: asuman.gedikbasi@istanbul.edu.tr; Tel.: 00905327009660

Abstract

Fibroblast growth factor 21 (FGF21), a pleiotropic hormone, is a significant modulator of energy homeostasis. We evaluated serum FGF21 levels in patients with a deficiency of mitochondrial aminoacyl t-RNA synthetase (aARSs). Six patients with mitochondrial aminoacyl tRNA synthetase deficiency and twelve healthy volunteers were included in this study. Whole-exome sequencing was used for molecular diagnosis. Serum FGF21 levels in the case group and healthy volunteers were analyzed using the enzyme-linked immunosorbent assay. Exome sequencing test revealed nine different pathogenic variants in the AARS2, EARS2, DARS2, SARS2, and WARS2 genes. A statistically significant difference was found between the serum FGF21 levels of the case and control groups: case group (n = 6), 882.49 ± 923.60 pg/mL; control group (n = 12), 20.89 ± 2.63 pg/mL ($p < 0.001$). The area under the ROC Curve for FGF21 in the differential diagnosis of mitochondrial aminoacyl-tRNA Synthetase Deficiency was 0.933 (0.786-0.962). Sensitivity and Specificity 100%, Positive and Negative Predicted Value 100% were found for FGF21 >27.4 pg/mL cut-off value. Assessment of FGF 21 levels as an indicator of mitochondrial damage in mt-aARS deficiency may provide insight into the level of damage. Investigation of the biochemical mechanisms underlying the different levels of damage caused by different aminoacyl tRNA synthetases will be important in terms of elucidating clinical heterogeneity.

Keywords: mitochondrial disease; mitochondrial aminoacyl tRNA synthetase deficiency; FGF21; AARS2; EARS2; DARS2; SARS2; WARS2

1. Introduction

The energy-producing organelles in cells, known as mitochondria, can be affected by a variety of physiological and genetic conditions in the body [1]. Mitochondria form dynamic networks that interact with other organelles, the cytoplasm, and each other to exchange chemicals and information. The fact that communication between mitochondria can cross cellular boundaries and even distinct tissues is becoming increasingly widely recognized. Many common conditions, including metabolic disorders, neurological diseases, and cancer, are associated with mitochondrial stress and the dysregulated mitochondrial unfolded protein response (UPRmt). Mitokines are essential inter-organ communication mediators that impact systemic metabolic and physiological processes. They are signaling molecules that are released by the mitochondrial stress response and UPRmt [2]. Genetic

and environmental factors leading to mitochondrial dysfunction can trigger a broader cellular stress response that releases mitokines such as fibroblast growth factor 21 (FGF21).

Mitochondrial diseases (MDs), caused by mutations in the nuclear and mitochondrial genomes, constitute an important group of rare inherited metabolic disorders. Rare diseases have been emphasized in recent years as they shed light on the pathophysiology of common diseases. Clinical and genetic heterogeneity make mitochondria-related rare disorders an ideal disease group for an untargeted sequencing approach in genetic diagnosis. Whole exome sequencing (WES) approaches have significantly improved diagnostic yield and improved the understanding of mitochondrial biology, opening potential therapeutic avenues [3]. Autosomal recessive aminoacyl-tRNA synthetases (ARS) deficiencies represent a group of rare inherited MDs with a severe course and multiple organ involvement. Mitochondrial ARS2 are a group of enzymes that catalyze the addition of amino acids to the newly synthesized polypeptide chain and play an important role in translation. They are the key enzymes responsible for loading amino acids of the same origin into tRNA, and this enzymatic pathway is the most important step in the conversion of genetic information into proteins of interest [4]. The nuclear genome contains 37 ARS genes: 17 in the cytoplasm (known as aARS1), 17 in the mitochondria (known as ARS2), and three encoding enzymes that act in both cellular compartments [4,5]. The first initial letter is the single letter symbol of the corresponding amino acid, and DARS2, EARS2, WARS2, SARS2, and AARS2 are rare MDs that are the prototypes of this group of mitochondrial aspartate t-RNA synthetase, glutamate t-RNA synthetase, tryptophan t-RNA synthetase, serine t-RNA synthetase, and alanine-tRNA synthetase mutations, respectively. The reason for the severe course and clinical heterogeneity is that ubiquitous enzymes involved in mitochondrial aminoacylation, and translation processes cannot be synthesised and function due to defects in mt-aARS2 [5,6]. Reduced or disrupted translation may cause impaired protein folding, and intracellular stress can increase the release of mitokines such as FGF21. It is mainly secreted by the liver, but it may also be expressed in skeletal muscle.

Comprehensive investigations conducted over the past few decades have demonstrated the critical role that FGF21 plays in controlling a wide range of essential metabolic pathways, such as the transmission of insulin signals, the development of muscles, and the reaction to stress in the muscles. FGF21 is believed to help cells and tissues recover their equilibrium after adapting to a stressful environment by coordinating these metabolic pathways [7]. It has been suggested that measuring the concentration of FGF21 in serum could be applied as a first-line diagnostic test to reduce the need for a muscle biopsy in the diagnosis of primary respiratory chain deficiencies affecting muscles in adults and children [8]. In our study, we aimed to evaluate serum FGF21 levels, a mitokine that has recently come to the forefront as an indicator of mitochondrial stress in patients with mt-aARSs deficiency.

2. Results

2.1. Study Groups

The demographic characteristics of six affected patients from six unrelated families and their molecular findings are given in Table 1a,b.

Table 1. a. The demographic characteristics, biochemical findings of patients.

Cases	Gender	Age (years)	Weight (kg)	Height (cm)	BMI Kg/m2	Lactate (mg/dL)	Pyruvate (mg/dL)	FGF21 (pg/mL)
C1	M	3.7	9.18	81.5	13.8	49.9	0.59	349,5
C2	M	8.1	20.1	120.5	13.8	45.7	0.75	266,61
C3	F	3.4	13.7	96	15.1	8.7	0.37	298.44
C4	M	15.3	46.5	162	17.7	19.8	0.45	233.8
C5	F	6.5	19.5	111.5	15.7	19.5	0.42	2029.45

C6	M	2.9	9.8	88	12.6	26	0.47	2117.14
----	---	-----	-----	----	------	----	------	---------

* M; male, F; female, BMI; Body mass index. Reference range of Lactate: 4.5-20 mg/dL and Pyruvate: 0.3-0.9mg/dL.

Table 1. b. The genetic characteristics and molecular findings of patients.

Cases	Parental consanguinity	Zygosity	Gene	Variant in nucleotide	Variant in peptide
C1	Yes	Hom	AARS2	c.302G>T	p.(Arg101His)
C2	No	Com.het	AARS2	c.277C>T	p.(Arg93Ter)
C3	No	Com.het	EARS2	c.845C>G	p.(Ser282Cys)
C4	No	Com.het	DARS2	c.319C>T	p.(Arg107Cys)
C5	Yes	Hom	SARS2	c.1283delC	p.(Pro428Leufs*3)
C6	Yes	Hom	WARS2	c.492+2T>C	p.(?)

* Hom.; homozygous; Com. het.; Compound heterozygous.

2.1.1. Clinical Findings

The clinical symptoms of the patients were highly heterogeneous. The patient with a pathogenic variant in the SARS2 gene presented with renal involvement, whereas developmental retardation and lactate elevation were predominant in patients with pathogenic variants in AARS2 and WARS2 genes. Headache and cranial involvement were the main complaints in patients with the DARS2 pathogenic variant. Mental/developmental retardation and severe cerebellar atrophy were present in the patient with a pathogenic variant in the EARS2 gene.

2.1.2. Laboratory Findings

The means, standard deviations, medians, and interquartile ranges for each of the continuous variables in participants were shown in Table 2. No statistically significant difference was observed between the mean age and gender distribution of the control and patient groups (p=0.743, p=0.732). Also, no statistically significant difference was observed between the mean Weight (kg), Height (cm) and BMI (p=0.682, p=0.815, p=0.896, respectively). Lactate (mg/dL), Pyruvate (mg/dl) and FGF21 (pg/mL) means of the patient group were found to be statistically significantly higher than the control group (p=0.001, p=0.015, p=0.001, respectively).

Table 2. Comparison of demographic characteristics and laboratory findings of patient and control groups.

		Control Group	Patient Group	p
Age (years)	Mean±SD	6,73±4,01	6,65±4,70	0,743†
	Median (IQR)	5,6 (3,55-8,13)	5,1 (3,28-9,9)	
Gender	Male (n, %)	7, %58,33	4, %66,67	0,732+
	Female (n, %)	5, %41,67	2, %33,33	
Weight (kg)	Mean±SD	20,33±13,09	19,8±13,88	0,682†
	Median (IQR)	16,55 (10,73-20,8)	16,6 (9,65-26,7)	
Height (cm)	Mean±SD	111,69±27,31	109,92±29,36	0,815†
	Median (IQR)	104,8 (90,5-124,73)	103,75 (86,38-130,88)	
BMI (kg/m2)	Mean±SD	14,91±2,03	14,78±1,80	0,896*

	Mean±SD	5,68±0,60	28,27±16,18	
Lactate (mg/dL)	Median (IQR)	5,75 (5,13-6,18)	22,9 (16,8-46,75)	0,001†
	Mean±SD	0,38±0,05	0,51±0,14	
Pyruvate (mg/dl)	Median (IQR)	0,375 (0,33-0,42)	0,46 (0,41-0,63)	0,015†
	Mean±SD	20,89±2,63	882,49±923,60	
FGF21 (pg/mL)	Median (IQR)	20,33 (19,01-22,24)	323,97 (258,41-2051,37)	0,001†

¹ SD; standard deviation, IQR; interquartile range, *Independent t test, †Mann Whitney U test †Fisher's exact test.

The area under the Receiver Operating Characteristic (ROC) curve was calculated to determine the efficiency of Lactate (mg/dL), Pyruvate (mg/dL), and FGF21 (pg/mL) variables in the differential diagnosis of mitochondrial aminoacyl-tRNA synthetase deficiency (Figure 1). The area under the ROC Curve values for Lactate (mg/dL), Pyruvate (mg/dL), FGF21 (pg/mL) in the differential diagnosis of Mitochondrial Aminoacyl-tRNA Synthetase Deficiency are presented in Table 3A and Sensitivity Specificity, Positive Predicted Value and Negative Predicted Value values for all three markers are presented in Table 3B.

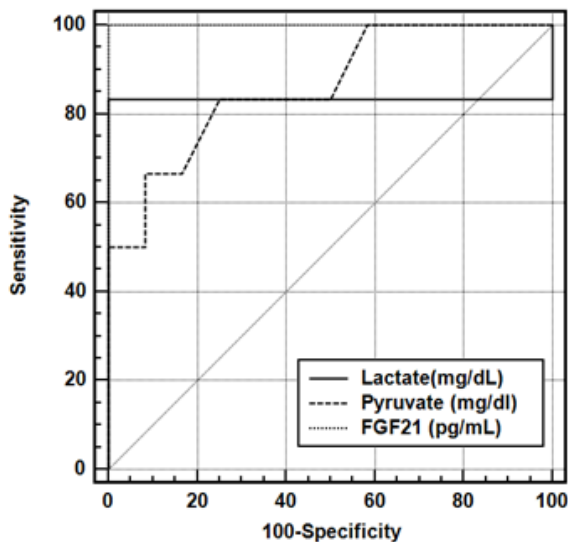


Figure 1. Area under the curve for biomarkers in the differential diagnosis of Mitochondrial Aminoacyl-tRNA Synthetase deficiency.

Table 3. Differential diagnosis of mitochondrial aminoacyl-tRNA synthetase deficiency. AUC for biomarkers (3A) and laboratory performance assessments (3B).

3A

	AUC	SE	95% CI
Lactate (mg/dL)	0,933	0,114	0,786-0,962
Pyruvate (mg/dl)	0,861	0,105	0,618-0,974
FGF-21 (pg/mL)	1,000	0,000	0,813-1,000

3B

	Criterion	Sensitivity	Specificity	PPV	NPV	LR (+)	LR (-)
Lactate (mg/dL)	>6,7 *	83,33	100,00	100,0	92,3		0,17

Pyruvate (mg/dl)	>0,41 *	83,33	75,00	62,5	90,0	3,33	0,22
FGF21 (pg/mL)	>27,4 *	100,00	100,00	100,0	100,0		0,00

After calculating the means and standard deviations of the total case and control levels, 2 patients were named as case group 2 (SARS2 and WARS2) because their FGF21 levels were much higher than the others. The other four patients were named case group 1. Case group 2 had higher FGF21 levels than case group 1 (2073.3 ± 43.8 and 309.4 ± 58.7 , $p > 0.05$). A violin plot was used to visualize the difference in FGF21 levels between groups, and since the difference in FGF21 levels between individuals was very high, its logarithm was taken (Figure 2).

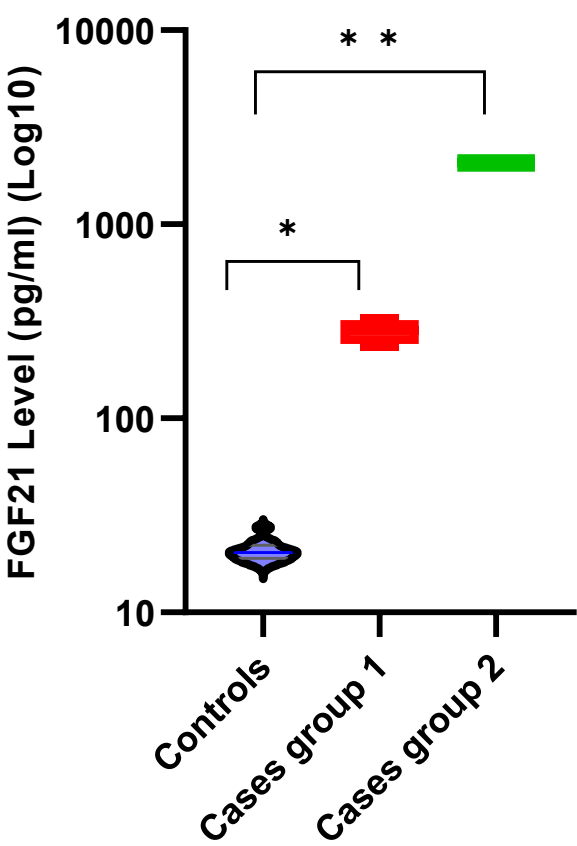


Figure 2. FGF-21 levels of case groups and control groups. * $p < 0.001$, ** $p < 0.05$ Cases group 1; AARS2 (n=2), EARS2, DARS2, Cases group 2; SARS2 and WARS2.

3. Discussion

In this study, we analyzed FGF21 levels in the serum of patients with MDs to assess whether it is a feasible biomarker for human mitochondria-related disorders. Serum FGF21 levels in patients with molecularly confirmed mitochondrial disease were statistically significantly higher than the control group, consistent with the literature. In the last decade, serum FGF21 levels have been proposed as a biomarker for mitochondrial diseases, and many studies have shown increased levels [9–15]. In a study involving a total of 140 participants (54 patients with mitochondrial disease, 20 adult patients with neuromuscular disease, and 66 healthy volunteers) to determine the validity and reliability of FGF21 as a biomarker, the serum levels were found to be more sensitive than the serum creatinine kinase, lactate, and pyruvate, which are classical biomarkers [9]. In the other study, a total of 13 biomarkers, including lactate, pyruvate, creatine kinase (CK), amino acid profiles, glutathione,

malondialdehyde, GDF-15, FGF21, gelsolin, neurofilament light chain, and circulating cell-free mtDNA, were comprehensively examined in MDs. GDF-15, followed by FGF21, has been shown to have the greatest value, although it is not perfect [15]. In our study, only two patients had increased lactate and pyruvate levels. FGF21 was high compared to control in all patients. This finding supported the fact that FGF21 was more sensitive. Subsequent studies have shown that serum FGF21 is a specific biomarker for caste defects of mitochondrial translation, including mitochondrial transfer-RNA mutations and primary and secondary mtDNA deletions. However, normal serum FGF21 does not rule out structural respiratory chain complex or mounting factor defects, which are important to be considered in diagnosis [16]. In another study, patients carrying mutations in nuclear DNA had higher FGF21 levels than patients with mtDNA mutations [17].

Aminoacyl-tRNA synthetases are critical enzymes responsible for attaching amino acids to their corresponding tRNA molecules, a pivotal step in translating genetic information into proteins. Each amino acid is paired with a distinct aminoacyl-tRNA synthetase. Mutations affecting tRNA synthetases alter the transport rate of the related amino acids, leading to clinical manifestations dependent on the levels and activity of the associated proteins. The extent of mitochondrial damage and clinical symptoms vary according to the synthesis of proteins linked to the affected amino acid [5,6]. MDs are caused by different molecular mechanisms and diagnostic cut-off values of FGF21 levels may vary. The strength of our study is that all included patients have similar molecular etiology. The limitation of our study is the small number of cases, but it is normal for rare diseases.

Biomarkers play a pivotal role in indicating the presence or absence of diseases, as well as monitoring their progression and treatment responses. Identifying a single dependable biomarker encompassing all MDs, particularly those involving primary mitochondrial respiratory chain dysfunction, remains exceedingly challenging. Instead, clinicians often rely on combinations of biomarkers, referred to as "biosignatures," rather than a solitary indicator for diagnostic purposes. While serum biomarkers like lactate and pyruvate find widespread use in diagnostics, they lack specificity for mitochondrial disorders. In certain cases, simultaneous assessments of creatine, plasma amino acids, and urinary organic acids can enhance the biosignature's utility [18].

In a recent study, FGF21 was identified as a potent metabolic regulator. This signaling protein belongs to the fibroblast growth factor family of endocrine substances and is secreted into the circulation. The liver, brown and white adipose tissue, are the main organs where FGF21 is expressed [19]. But the liver is the primary source, and its secretion is induced by nutritional and cellular stress signals. Hepatic FGF21 is produced in response to nutritional stressors such as prolonged fasting, a ketogenic diet, amino acid deprivation, or simple sugar consumption [20]. In our study, we did not identify any significant correlation between high FGF21 levels and the body mass index (BMI) or age of our patients.

In our study, FGF21 levels were significantly higher in patients with different aminoacyl t-RNA synthase deficiencies compared with the control group. It was especially higher in SARS2 and WARS2 patients (Figure 2). This supports the idea that the degree of mitochondrial damage and clinical findings vary according to the amount of synthesis of related amino acid-related proteins. In this study, we aim to investigate the importance of this biomarker in the diagnosis of MDs caused by different mt-aARS deficiencies and shed light on the ongoing evaluation of various approaches in this field.

4. Materials and Methods

4.1. Study Design and Participants

This study was carried out with six probands who were followed up in the Department of Pediatric Metabolism of the Istanbul University Faculty of Medicine and whose molecular diagnosis was finalized by detecting mutations in the aARS genes. The control group consisted of 12 healthy volunteers. Exome sequencing revealed nine different pathogenic variants in five genes encoding aminoacyl-tRNA synthetase (AARS2, EARS2, DARS2, SARS2, and WARS2). Sanger sequencing was

performed for the segregation of pathogenic alterations. The study protocol was approved by the local ethics committee of our hospital. After a detailed explanation of the aims and scope of the study in accordance with the principles of the World Medical Association Declaration of Helsinki, a consent form was obtained from the patient's parents.

4.2. Biochemical Analysis

All patients were in a stable phase during the phlebotomy. After fasting for approximately 4 hours, the blood sample taken into a gel tube (BD, USA) was left for 45 minutes and centrifuged at 4000 rpm for 10 minutes. The supernatant was stored at -80 °C until the time of analysis. The analysis was performed by enzyme-linked immunosorbent assay (ELISA) after all samples were collected. Serum FGF21 measurements (Catalogue No. DF2100, Quantikine ELISA Kit, R&D Systems, Inc., Minneapolis, USA) were performed using the sandwich enzyme immunoassay technique, and results were evaluated in pg/mL. The assay is based on the method of quantitative sandwich enzyme immunoassay, and the intra-assay and inter-assay coefficients of variation (CV%) were 2.9–3.9% and 5.2–10.9%, respectively. The traditional biomarkers lactate (LA) and pyruvate (PA) were evaluated in plasma together with FGF21. Plasma lactate levels were analyzed with colorimetric method in the ADVIA Chemistry LAC original reagent in the ADVIA Chemistry XPT autoanalyzer (Siemens Healthcare Diagnostics, Germany). Plasma pyruvate levels were measured spectrophotometrically with a commercial kit according to the manufacturer's instructions (Ben S.r.l. Biochemical Enterprise, Italy). Pyruvate and lactate results were expressed as mg/dL. Reference ranges for lactate and pyruvate are 4.5–20 mg/dL and 0.3–0.9 mg/dL, respectively.

4.3. Statistical Analysis

This study performed statistical analyses using the NCSS (Number Cruncher Statistical System) 2007 Statistical Software (Utah, USA) package program. In addition to descriptive statistical methods (mean, standard deviation, median, interquartile range), the Shapiro-Wilk normality test was used to evaluate the distribution of variables, the independent t-test was used to compare paired groups of variables with normal distribution, Mann Whitney U test was used to compare paired groups of variables that did not show normal distribution, Fisher exact test was used to compare qualitative data. The results were evaluated at a significance level of $p < 0.05$.

5. Conclusions

Assessment of FGF21 levels as an indicator of mitochondrial damage in mt-aARS deficiency may provide insight into the level of damage. Investigation of the biochemical mechanisms underlying the different levels of damage caused by different aminoacyl tRNA synthetases will be important in terms of elucidating clinical heterogeneity. This study also sheds light on the use of FGF21 as a biomarker for the demonstration of secondary mitochondrial damage in chronic diseases.

Author Contributions: Conceptualization: Tekin Neijmann S; Gokcay GF; Gedikbasi A Methodology: Tekin Neijmann S; Gunes D; Karaca M; Balci MC; Karaman V; Gokcay GF; Gedikbasi A. Investigation: Tekin Neijmann S; Gunes D; Karaca M; Balci MC; Karaman V; Gedikbasi A, Visualization: Karaman V; Gedikbasi A, Funding acquisition: Gedikbasi A., Project administration: Tekin Neijmann S; Gedikbasi A, Supervision: Gokcay GF , Writing – original draft: Tekin Neijmann S, Writing – review & editing: Tekin Neijmann S; Gunes D; Karaca M; Balci MC; Karaman V; Gokcay GF; Gedikbasi A.

Funding: This study was funded by the Scientific Research Projects Coordination Unit of Istanbul University, Project No: TSA-2024-40454.

Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki, and the studies involving human participants were reviewed and approved by the Faculty of Medicine Ethics Council of Istanbul University (08.06.2018/11).

Informed Consent Statement: Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

Data Availability Statement: The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found at: <https://www.ncbi.nlm.nih.gov/snp/>, SCV001423134.1 <https://www.ncbi.nlm.nih.gov/snp/>, SCV001423135.1 <https://www.ncbi.nlm.nih.gov/snp/>, SCV001571651.1 <https://www.ncbi.nlm.nih.gov/snp/>, SCV001571652.1.

Acknowledgments: We would like to thank Rana Konyalioglu for her efforts in statistical analyses. The authors also thank the patients and their families for participating in this study.

Conflicts of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

References

1. Sahin, E.; Alver, A. Mitohormesis and Regulatory Mechanisms. *Farabi Med. J.* **2020**, *1*, 21–26.
2. Zhang, B.; Chang, J.Y.; Lee, M.H.; Ju, S.H.; Yi, H.S.; Shong, M. Mitochondrial Stress and Mitokines: Therapeutic Perspectives for the Treatment of Metabolic Diseases. *Diabetes Metab. J.* **2024**, *48*, 1–18.
3. Gedikbasi, A.; Toksoy, G.; Karaca, M.; Gulec, C.; Balci, M.C.; Uyguner, Z.O. Clinical and Bi-Genomic DNA Findings of Patients Suspected to Have Mitochondrial Diseases. *Front. Genet.* **2023**, *14*, 1191159.
4. Quirós, P.M.; Mottis, A.; Auwerx, J. Mitonuclear Communication in Homeostasis and Stress. *Nat. Rev. Mol. Cell Biol.* **2016**, *17*, 213–226.
5. Felhi, R.; Charif, M.; Sfaihi, L.; Mkaouar-Rebai, E.; Desquiere-Dumas, V.; Kallel, R.; et al. Mutations in aARS Genes Revealed by Targeted Next-Generation Sequencing in Patients with Mitochondrial Diseases. *Mol. Biol. Rep.* **2020**, *47*, 3779–3787.
6. Fine, A.S.; Nemeth, C.L.; Kaufman, M.L.; Fatemi, A. Mitochondrial Aminoacyl-tRNA Synthetase Disorders: An Emerging Group of Developmental Disorders of Myelination. *J. Neurodev. Disord.* **2019**, *11*, 29.
7. Li, S.; Chen, J.; Wei, P.; Zou, T.; You, J. Fibroblast Growth Factor 21: A Fascinating Perspective on the Regulation of Muscle Metabolism. *Int. J. Mol. Sci.* **2023**, *24*, 16951.
8. Suomalainen, A.; Elo, J.M.; Pietiläinen, K.H.; Hakonen, A.H.; Sevastianova, K.; Tyynismaa, H. FGF-21 as a Biomarker for Muscle-Manifesting Mitochondrial Respiratory Chain Deficiencies: A Diagnostic Study. *Lancet Neurol.* **2011**, *10*, 806–818.
9. Turnbull, D. A New Biomarker for Mitochondrial Disease. *Lancet Neurol.* **2011**, *10*, 777–778.
10. Davis, R.L.; Liang, C.; Edema-Hildebrand, F.; Riley, C.; Needham, M.; Sue, C.M. Fibroblast Growth Factor 21 Is a Sensitive Biomarker of Mitochondrial Disease. *Neurology* **2013**, *81*, 1819–1826.
11. Davis, R.L.; Liang, C.; Sue, C.M. A Comparison of Current Serum Biomarkers as Diagnostic Indicators of Mitochondrial Diseases. *Neurology* **2016**, *86*, 2010–2015.
12. Morovat, A.; Weerasinghe, G.; Nesbitt, V.; Hofer, M.; Agnew, T.; Poulton, J. Use of FGF-21 as a Biomarker of Mitochondrial Disease in Clinical Practice. *J. Clin. Med.* **2017**, *6*, 80.
13. Scholle, L.M.; Lehmann, D.; Deschauer, M.; Kraya, T.; Zierz, S. FGF-21 as a Potential Biomarker for Mitochondrial Diseases. *Curr. Med. Chem.* **2018**, *25*, 2070–2081.
14. Watson, E.; Davis, R.; Sue, C.M. New Diagnostic Pathways for Mitochondrial Disease. *J. Transl. Genet. Genom.* **2020**, *4*, 188–202.
15. Shayota, B.J. Biomarkers of Mitochondrial Disorders. *Neurotherapeutics* **2024**, *21*, e00325.
16. Lehtonen, J.M.; Forsström, S.; Bottani, E.; Viscomi, C.; Baris, O.R.; Suomalainen, A. FGF21 Is a Biomarker for Mitochondrial Translation and mtDNA Maintenance Disorders. *Neurology* **2016**, *87*, 2290–2299.

17. Wesół-Kucharska, D.; Rokicki, D.; Greczan, M.; Kaczor, M.; Czekuć-Kryśkiewicz, E.; Piekutowska-Abramczuk, D. The Fibroblast Growth Factor 21 Concentration in Children with Mitochondrial Disease Does Not Depend on the Disease Stage, but Rather on the Disease Genotype. *Pediatr. Endocrinol. Diabetes Metab.* **2022**, *28*, 141–151.
18. Burtscher, J.; Soltany, A.; Visavadiya, N.P.; Burtscher, M.; Millet, G.P.; Khoramipour, K.; et al. Mitochondrial Stress and Mitokines in Aging. *Aging Cell* **2023**, *22*, e13770.
19. Domouzoglou, E.M.; Vlahos, A.P.; Vasileios, K.; Cholevas, V.K.; Michail, I.; Papafaklis, M.I.; Chaliasos, N.; Siomou, E. Association of Fibroblast Growth Factor 21 with Metabolic Syndrome and Endothelial Function in Children: A Prospective Cross-Sectional Study on Novel Biomarkers. *Ann. Pediatr. Endocrinol. Metab.* **2021**, *26*, 242–251.
20. Szczepańska, E.; Gietka-Czernel, M. FGF21: A Novel Regulator of Glucose and Lipid Metabolism and Whole-Body Energy Balance. *Horm. Metab. Res.* **2022**, *54*, 203–211.

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.