Таблица 9

Изменения, протекающие в рядах Гисса при посеве суспензий E. coli – инкубация 72 часа

Мальтоза	Глюкоза	Маннитол	Сорбитол	Лактоза
E. coli (+)	E. coli (+)	E. coli (+) Г	E. coli (+)	E. coli (+) Г
E. coli c Se (+)	E. coli c Se (+)	E. coli c Se (+)(-)	E. coli c Se (+)	E. coli c Se (+) Г
E. coli c Zn и Cr (+)	E. coli c Zn и Cr (+)	E. coli c Zn и Cr (+) Г	E. coli c Zn и Cr (+)	E. coli c Zn и Cr (+) Г

Таблица 10

Изменения сред Гисса при посеве суспензий со St. aureus – инкубация 72 часа

Мальтоза	Глюкоза	Маннитол	Сорбитол	Лактоза
E. coli (+)	E. coli (+)	E. coli (+) Г	E. coli (+)	E. coli (+) Г
E. coli c Se (+)	E. coli c Se (+)	E. coli c Se (+)(-)	E. coli c Se (+)	E. coli c Se (+) Г
E. coli c Zn и Cr (+)	E. coli c Zn и Cr (+)	E. coli c Zn и Cr (+) Г	E. coli c Zn и Cr (+)	E. coli c Zn и Cr (+) Г

Таким образом изучение метаболической активности микроорганизмов по определению их гликолитической активности на средах Гисса показало, что сахаролитические свойства с образованием кислот и газа ярко выражены для E. coli как с добавлением Se, так и Zn с Cr, на всех интервалах инкубации. В то время как гликолитические процессы для St. aureus менее выражены и отсутствует процесс газообразования.

Сравнительный анализ метаболической активности микроорганизмов без добавления и с добавлением наночастиц Se и Zn с Cr показал, что метаболические процессы с образованием кислот и газа более выражены у E. coli, по сравнению со St. aureus как без наночастиц Se и Zn с Cr, так и с их добавлением, что свидетельствует о более глубоких ферментативных процессах, происходящих под воздействием на наночастиц.

ЛИТЕРАТУРА

1. Скоринова К.Д., Кузьменко В.В., Василенко А.И. Перспектива создания лекарственных препаратов на основе наночастиц селена (обзор). *Разработка и регистрация лекарственных средств*. 2020;9(2):33-44.

2. Liu H, Sun Y, Ramos KR, Nisola GM, Valdehuesa KN, Lee WK, Park SJ, Chung WJ. Combination of Entner-Doudoroff pathway with MEP increases isoprene production in engineered Escherichia coli. PLoS One. 2013 Dec 20;8(12)

3. Capra EJ, Laub MT. Evolution of two-component signal transduction systems. Annu Rev Microbiol. 2012;66:325-47. doi: 10.1146/annurev-micro-092611-150039. Epub 2012 Jun 28.

MITOCHONDRIAL DNA VARIANT M.4344T>C IN $_{T}$RNAGln CAUSES DEVELOPMENTAL DELAY

МУТАЦИЯ МИТОХОНДРИАЛЬНОЙ ДНК M.4344T>C $_{T}$RNAGln ВЫЗЫВАЕТ ОТСТАВАНИЕ В РАЗВИТИИ

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The current work discusses the identification of a novel pathogenic mitochondrial DNA variant, m.4344T>C, in tRNAGln and its implications for patients with developmental delay. The variant was identified in an 18-months-old boy with developmental delay, and it was found to be heteroplasmic in the patient's blood and oral epithelial cells. Functional studies demonstrated that the m.4344T>C variant impaired mitochondrial complexes I, III, and IV contents, resulting in defective mitochondrial respiration, elevated mitochondrial ROS production, reduced mitochondrial membrane potential, and decreased mitochondrial ATP levels compared to wild-type cybrids. This study expanded the genetic variant spectrum of mitochondrial diseases and provided a better understanding of the phenotypes associated with mitochondrial tRNAGln gene mutations, contributing to the clinical diagnosis of developmental delay in patients with mitochondrial DNA variants.

В настоящей работе обсуждается идентификация нового патогенного варианта митохондриальной ДНК, m.4344T>C, в tRNAGln и его значение для пациентов с задержкой развития. Вариант был идентифицирован у 18-месячного мальчика с задержкой развития и оказался гетероплазматическим в крови пациента и эпителиальных клетках полости рта. Функциональные исследования показали, что вариант m.4344T>C нарушает содержание митохондриальных комплексов I, III и IV, что приводит к нарушению митохондриального дыхания, повышению продукции митохондриальных АФК, снижению потенциала митохондриальной мембраны и снижению уровня митохондриального АТФ по сравнению с цибридами дикого типа. Это исследование расширило спектр генетических вариантов митохондриальных заболеваний и позволило лучше понять фенотипы, связанные с мутациями гена митохондриальной тРНКGln, что способствовало клинической диагностике задержки развития у пациентов с вариантами митохондриальной ДНК.

Keywords: мутации митохондриальной тРНК, комплексы дыхательной цепи, функция митохондрий, митохондриальные заболевания, гибридные клетки, скорость потребления кислорода.

Ключевые слова: mitochondrial mRNA mutations, respiratory chain complexes, mitochondrial function, mitochondrial diseases, hybrid cells, oxygen consumption rate.

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The prevalence of mitochondrial diseases in the general population is estimated to be approximately 1 in 5,000 individuals [1]. Mitochondrial diseases are a group of genetic disorders caused by mutations in mitochondrial DNA and nuclear DNA. These mutations can lead to defects in the mitochondrial oxidative phosphorylation (OXPHOS) system, impacting various cellular functions and energy production processes [2]. Despite advancements in genetic testing and sequencing technologies, many patients with mitochondrial diseases remain genetically undiagnosed, indicating that there are likely undiscovered genetic mutations contributing to the disease.

The relationship between genetic variants and developmental delay, particularly in the context of mitochondrial diseases, is crucial for understanding the underlying mechanisms of this condition. Genetic mutations in mitochondrial DNA and nuclear DNA can lead to mitochondrial dysfunction, affecting various cellular processes and energy production (fig. 1) [3-5]. In the case of the m.4344T>C variant identified in mitochondrial tRNAGln from a patient with developmental delay, this mutation impairs mitochondrial function by decreasing the contents of complexes I, III, and IV. This impairment results in defective mitochondrial respiration, elevated ROS production, reduced mitochondrial membrane potential, and decreased ATP levels compared to wild-type cybrids. The genetic spectrum of mitochondrial diseases is diverse, with mutations in genes encoding proteins involved in the oxidative phosphorylation (OXPHOS) system being commonly associated with developmental delay.

The OXPHOS system plays a critical role in cellular energy production, and disruptions in this system due to genetic variants can lead to developmental delays and other clinical manifestations of mitochondrial diseases.Overall, genetic variants like the m.4344T>C mutation can directly impact mitochondrial function, leading to developmental delay through mechanisms such as impaired energy production, oxidative stress, and compromised cellular processes. Understanding the relationship between specific genetic variants and developmental delay is essential for advancing diagnostic methods and potential therapeutic interventions for individuals affected by mitochondrial diseases.

The m.4344T>C variant is located in the anticodon stem of the tRNAGln gene, leading to a disruption in the structure and function of the tRNA molecule. This alteration can affect the efficiency of mitochondrial translation, leading to impaired protein synthesis in the mitochondria. The reduced levels of functional mitochondrial proteins can disrupt cellular energy production and overall mitochondrial function, contributing to developmental delays.

Individuals harboring the m.4344T>C variant often present with a spectrum of clinical symptoms, with developmental delay being a prominent feature. Other common manifestations may include neurological deficits, muscle weakness, and metabolic abnormalities. The severity and onset of symptoms can vary among affected individuals, highlighting the heterogeneity of mitochondrial disorders associated with this variant.

Diagnosing developmental delays associated with the m.4344T>C variant can be challenging due to the variability in clinical presentation and the overlap of symptoms with other mitochondrial disorders. Genetic testing, including sequencing of the mitochondrial genome, is crucial for identifying the specific mtDNA variant. Additionally, functional studies to assess the impact of the variant on tRNAGln function can provide valuable insights into the pathogenicity of the mutation.

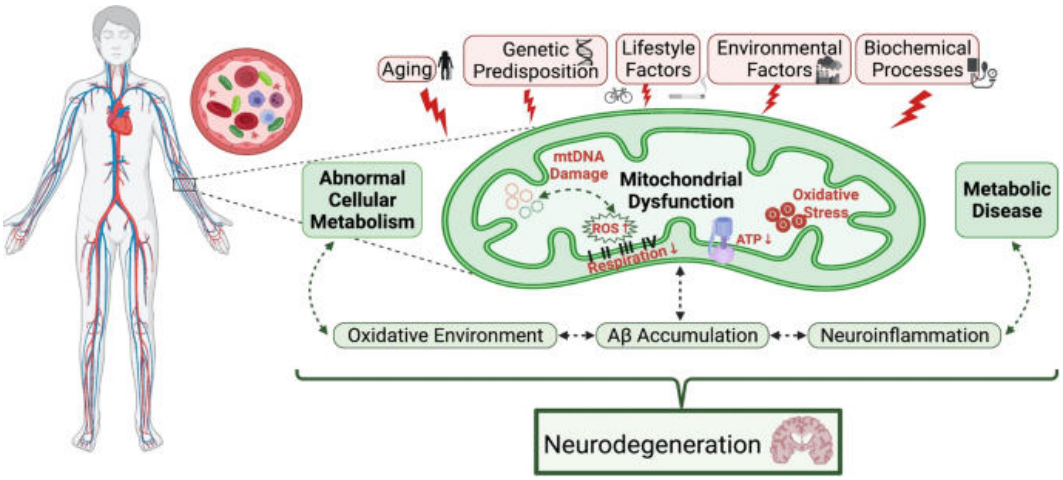

Figure 1 – Graphical overview of global working hypothesis for risk factors and cellular/molecular processes that contribute to neurodegeneration

Currently, treatment options for developmental delays caused by the m.4344T>C variant are limited. Management primarily focuses on supportive care to address the symptoms and optimize the overall well-being of affected individuals. Research efforts are ongoing to explore potential therapeutic interventions targeting mitochondrial function and translation efficiency to alleviate the clinical manifestations associated with this pathogenic variant.

The methods used in the study to determine the impact of the m.4344T>C variant on mitochondrial function included the following:

1. Whole exome sequencing (WES) and mitochondrial genomic sequencing using Illumina HiSeq 2000 sequencer to identify the pathogenic variants.
2. Collection of serum samples from the proband and his parents for next generation sequencing to identify the pathogenic variants.
3. Generation of cybrid cells to perform functional assays.
4. Measurement of mitochondrial ATP levels to assess the impact of the mutation on ATP production.
5. Evaluation of mitochondrial membrane potential to determine the effect of the variant on membrane potential.
6. Assessment of mitochondrial ROS production to understand the impact of the variant on reactive oxygen species levels.
7. Analysis of mitochondrial respiratory function, including oxygen respiration, OXPHOS complexes contents, and basal respiration to evaluate the overall impact on mitochondrial function.

These methods collectively provided a comprehensive assessment of the impact of the m.4344T>C variant on mitochondrial function (fig.2).

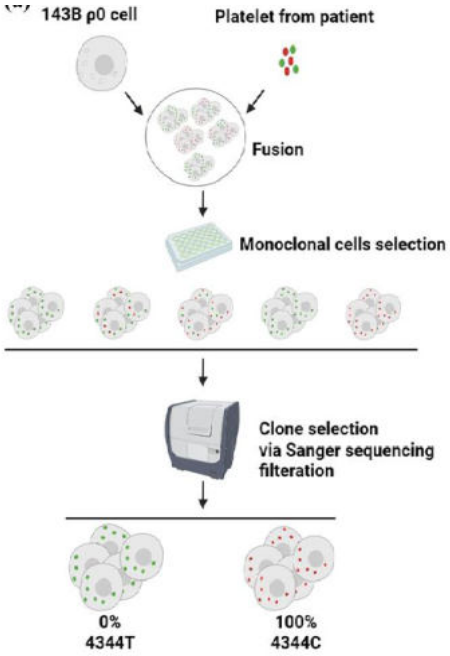

Figure 2 – Schematic diagram of monoclonal selection. Different 230 colors indicate different mutations in the mtDNA

The study found that the m.4344T>C variant impaired mitochondrial function in several ways:

1. The mutant variant decreased the contents of mitochondrial complexes I, III, and IV, leading to defective mitochondrial respiration.

2. It resulted in elevated mitochondrial ROS production, indicating increased oxidative stress levels.

3. The variant led to a reduction in mitochondrial membrane potential.

4. Mitochondrial ATP levels were significantly decreased in cells with the mutant variant compared to wild-type cells, suggesting a decrease in cellular ATP levels.

5. The impaired mitochondrial respiratory function caused an increase in electron leakage, further contributing to elevated oxidative stress levels.

6. Mutant cybrid cells exhibited elevated mitochondrial ROS levels, reduced mitochondrial membrane potential, and decreased cellular ATP levels compared to wild-type cybrid cells, indicating severe impairment of mitochondrial OXPHOS function.

The m.4344T>C variant identified in mitochondrial tRNAGln from a patient with developmental delay was found to significantly impact mitochondrial function by decreasing the contents of complexes I, III, and IV. This impairment led to defective mitochondrial respiration, elevated ROS production, reduced membrane potential, and decreased ATP levels compared to wild-type cybrids. The study expands the genetic variant spectrum of mitochondrial diseases and enhances the understanding of phenotypes associated with mutations in the mitochondrial tRNAGln gene. These findings contribute to the clinical diagnosis and management of mitochondrial diseases by providing insights into the genetic basis of developmental delays linked to mitochondrial dysfunction.

The m.4344T>C variant identified in mitochondrial tRNAGln from a patient with developmental delay holds significant importance in the context of mitochondrial diseases. This variant was found to impair mitochondrial function by decreasing the contents of complexes I, III, and IV, leading to defective mitochondrial respiration, elevated ROS production, reduced membrane potential, and decreased ATP levels compared to wild-type cybrids. The mutant loads of m.4344T>C were notably high in the patient's blood and oral epithelial cells. Furthermore, multialignment analysis revealed the high evolutionary conservation of this nucleotide. The significance of the m.4344T>C variant lies in its contribution to expanding the genetic variant spectrum of mitochondrial diseases. By elucidating the impact of this variant on mitochondrial function and cellular processes, the study enhances the understanding of phenotypes associated with mutations in the mitochondrial tRNAGln gene. This knowledge is crucial for improving the clinical diagnosis and management of mitochondrial diseases linked to mitochondrial dysfunction. Overall, the identification and characterization of the m.4344T>C variant provide valuable insights into the genetic mechanisms underlying developmental delays associated with mitochondrial tRNAGln gene mutations, thereby advancing our understanding of mitochondrial diseases.

Functional studies demonstrated that the m.4344T>C variant affects the stability of the tertiary structure of tRNAGln, leading to inefficient protein translation. This impairment in protein synthesis within the mitochondria results in compromised mitochondrial function, affecting energy production and cellular processes. Additionally, the high evolutionary conservation of this nucleotide suggests its critical role in maintaining normal mitochondrial function. Overall, the m.4344T>C variant disrupts mitochondrial complexes and impairs essential cellular processes, ultimately contributing to developmental delay in affected individuals. By elucidating these mechanisms, this research enhances our understanding of how specific genetic variants can lead to mitochondrial dysfunction and associated clinical manifestations.

The relationship between mitochondrial membrane potential and developmental delay is significant in the context of mitochondrial diseases. The m.4344T>C variant identified in mitochondrial tRNAGln from a patient with developmental delay was found to decrease mitochondrial membrane potential compared to wild-type cybrids. This reduction in membrane potential is a crucial aspect of mitochondrial dysfunction associated with this variant. Mitochondrial membrane potential plays a vital role in cellular energy production and overall mitochondrial function. A decrease in membrane potential, as observed in individuals with the m.4344T>C variant, can lead to impaired ATP production, disrupted cellular processes, and increased oxidative stress due to elevated ROS production. These effects contribute to the pathogenesis of developmental delay seen in patients with mitochondrial diseases linked to this specific genetic variant.Therefore, the relationship between mitochondrial membrane potential and developmental delay lies in the impact of reduced membrane potential on cellular energy metabolism, oxidative stress levels, and overall mitochondrial function. Understanding and addressing these disruptions are crucial for elucidating the mechanisms underlying developmental delays associated with mitochondrial dysfunction caused by genetic variants like m.4344T>C.

Further research is warranted to elucidate the intricate molecular mechanisms underlying the pathogenicity of the m.4344T>C variant in tRNAGln and its specific role in developmental delays. Investigating potential therapeutic targets and personalized treatment approaches tailored to mitochondrial disorders associated with this variant holds promise for improving clinical outcomes and quality of life for affected individuals.

In conclusion, the pathogenic mitochondrial DNA variant m.4344T>C in tRNAGln is a significant contributor to developmental delays, emphasizing the importance of comprehensive genetic and functional analyses in the diagnosis and management of mitochondrial disorders. Continued research efforts are essential for advancing our understanding of this variant and developing targeted interventions to address the complex clinical manifestations associated with it.

REFERENCES

1. Schon KR, Ratnaike T, van den Ameele J, Horvath R, Chinnery PF. Mitochondrial diseases: a diagnostic revolution // Trends in Genetics. 2020. 36(9). 702-717.

2. Pello R, Martín MA, Carelli V, Nijtmans LG, Achilli A, Pala M, Torroni A, Gomez-Durán A, Ruiz-Pesini E, Martinuzzi A. Mitochondrial DNA background modulates the assembly kinetics of OXPHOS complexes in a cellular model of mitochondrial disease // Human molecular genetics. 2008. 17(24). 4001-4011.

3. Liu G, Shen X, Sun Y, Lv Q, Li Y, Du A. Heteroplasmy and phenotype spectrum of the mitochondrial tRNALeu (UUR) gene m.3243A>G mutation in seven Han Chinese families // Journal of the Neurological Sciences. 2020. 408. 116562.

4. Nesbitt V, Pitceathly RD, Turnbull DM, Taylor RW, Sweeney MG, Mudanohwo EE, Rahman S, Hanna MG, McFarland R. The UK MRC Mitochondrial Disease Patient Cohort Study: clinical phenotypes associated with the m. 3243A>G mutation–implications for diagnosis and management // Journal of Neurology, Neurosurgery & Psychiatry. 2013. 84(8). 936-938.

5. Majamaa K, Moilanen JS, Uimonen S, Remes AM, Salmela PI, Kärppä M, Majamaa-Voltti KA, Rusanen H, Sorri M, Peuhkurinen KJ. Epidemiology of A3243G, the mutation for mitochondrial encephalomyopathy, lactic acidosis, and strokelike episodes: prevalence of the mutation in an adult population // The American Journal of Human Genetics. 1998. 63(2). 447-454.

КВАНТОВО-ХИМИЧЕСКОЕ МОДЕЛИРОВАНИЕ И ОЦЕНКА БИОЛОГИЧЕСКОЙ АКТИВНОСТИ 2,4-ДИ-ТЕРТ-БУТИЛ-6-МОРФОЛИНОФЕНОЛА ПРОТИВ ВИЧ ПЕРВОГО ТИПА

QUANTUM CHEMICAL MODELING AND EVALUATION OF THE BIOLOGICAL ACTIVITY OF 2,4-DI-TERT-BUTYL-6-MORPHOLINOPHENOL AGAINST TYPE 1 HIV

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В работе представлено новое производное морфолина: 2,4-ди-терт-бутил-6-морфолинофенол, которое может служить потенциальной лекарственной субстанцией против ВИЧ 1 типа. Проведена оценка его биологической активности с помощью программного пакета Gaussian09W методом B3LYP/Midix. При проведении молекулярного докинга выявлено образование термодинамически устойчивого комплекса между белком, отвечающим за заболевание ВИЧ-1 с 2,4-ди-терт-бутил-6-морфолинофенолом.

This research paper presents a new morpholine derivative that can serve as a potential drug against type 1 HIV. The biological activity of the selected compound was evaluated and the density functional theory was calculated using the Gaussian09W B3LYP/Midix quantum kit. Molecular docking revealed the binding between the protein responsible for HIV-1 disease and the studied morpholine derivative and demonstrated excellent binding affinity.

Ключевые слова: ВИЧ-1 типа, докинг, производное морфолина

Keywords: HIV-1 type, docking, morpholine derivative

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Вирус иммунодефицита человека (ВИЧ) – одноцепочечный РНК-вирус, имеющий 2 подтипа: ВИЧ-1 и ВИЧ-2. Подтип ВИЧ-1 связан с патогенезом и прогрессированием синдрома приобретенного иммунодефицита (СПИД).

За последние десятилетия произошел значительный прогресс в лечении ВИЧ. Демографическая ситуация среди людей, живущих с ВИЧ, изменилась в сторону старения населения, особенно в разных странах. Этот тренд может быть объяснен разработкой новых классов антиретровирусных препаратов (АРВ). Такие препараты характеризуются высокой эффективностью, лучшей переносимостью и в большинстве случаев имеют меньшее количество взаимодействий с другими лекарствами [1].

Большинство лекарственных препаратов содержат гетероциклы, так как они обладают донорами и акцепторами водородных связей и могут взаимодействовать с ферментами-мишенями и рецепторами через водород-