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Brief Report

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The Mitochondrial DNA D-Loop Variations in Depressed Patients

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Abstract

Depression is one of an important health problems in the last years, the predisposition to the depression still under investigations, the present study deal with the mtDNA variation loci in depression patients compare with control, PCR-sequencing was used to determine mutations, results show that there were among 173 nucleotides of the D-loop (16034-16207) there were 19 sites differentiated in non-significant differences, about 8 types of substitution mutations were observed included (A>C, A>G, T>C, G>A, C>T, C>A, C>G, G>A), insertion and deletion mutations also founded, non-significant differences were appeared in the types and number of mutations in this segment of mtDNA. The present output concluded that there wasn't association between D-loop variations and depression disease.

Keywords: Mitochondrial DNA • D-loop • Variations • Depression patients

Introduction

The Mitochondria organelle is playing a critical role in energy production, lipid and steroid metabolism, and cellular stability, as well as modulating Ca^{2+} levels, maintaining ROS levels, and regulating apoptosis [1]. As a result, mitochondrial dysfunction not only makes it difficult for cells to get their energy needs, but it may also be contributed in the neuronal communication and cellular signals impairment [2].

The notion of neuroplasticity is at the center of a new mood disorder theory, which focuses on bipolar disorder and depression. The term "neuroplasticity" refers to the brain's malleability, which includes synaptic and non-synaptic plasticity; Synaptic plasticity is the process of neurons responding to changes in their exogenous and endogenous environment by generating alterations in brain pathways and synapses. It includes synaptic-genesis, axon and dendritic development, and the elimination of superfluous linkage between neurons. Mitochondria play a crucial role in neuroplasticity, and it has been well established that stress causes structural and functional damage in many brain areas of individuals suffered from depression, resulting in decreased neuroplasticity [3].

The Major Depressive Disorder (MDD) is a prevalent mental condition [4]. Some investigations suggested that the neurotropic factors, neuroplasticity, and mitochondrial dysfunction for biological hypotheses proposed of MDD because the MDD pathophysiology still under studying [5]. the impairment of Mitochondrial functions was affecting in cellular processes due to impairments in cellular resilience and synaptic plasticity [6], and has also been associated with psychiatric illnesses, such as bipolar disorder, MDD, anxiety disorder and schizophrenia [7,8].

Several investigations have shown a connection of depression with mitochondrial malfunction, and systematic psychiatric assessments of individuals with mitochondrial illness have revealed that around half of these patients had a lifetime MDD prevalence [9,10]. Measuring mtDNA copy numbers can be used to measure mitochondrial function indirectly. Cells that require a lot of energy, including heart cells, skeletal muscle cells, and neurons, need a lot of ATPS and have a lot of mtDNA copies, which is considered a measure of mitochondrial energy function [11]. The presence of abnormal mtDNA copy numbers has been linked to a variety of mental illnesses [12].

Materials and Methods

Sample collection

A 24 depression, blood samples were collected from (Margan medical city Hospital, Babylon–Iraq), which were diagnosed by specialist, physician prof. Dr. kareem Naser, and 20 blood samples of control were collected from healthy people, their age ranged (23 years to 70 years).

Genomic DNA extraction

DNA was isolated from whole blood for both groups using a DNA extraction kit (Favorgen), and then DNA concentrations and purity were estimated using a Nanodrop.

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The primer of HV1a gene

It was used Forward 5- CAC CAT TAG CAC CCA AAG CT-3, Reverse 5- GGC TTT GGA GTT GCA GTT GAT -3 the PCR amplicon size (280 bp) with annealing temperature reached to 58.8ºC The products visualized by electrophoresis (1agarose, 75 V, 20 mA for 40 min).

Capillary electrophoresis

The capillary electrophoresis was carried out using genetic analyzer (applied bio system 3600), data were interpretated using software, then analysis by NCBI blast, https://blast.ncbi.nlm.nih.gov/Blast.cgi, sms bioinformatics software http://www.bioinformatics.org/sms2/pcr_products.html, and MAFFT version 7 https://mafft.cbrc.jp/alignment/server/. In comparison with (15978- 16257 in Homo sapiens mitochondrion).

Results

Twenty-four patients were diagnosed with major depression by physician, The age of the patients (16 females and 8 males) was in the range of 23 years to 70 years, and twenty samples as control group age ranged (20 years to 62 years, DNA was extracted from the whole blood of patients and control, its concentration was (50 ng/µl to 150 ng/µl) and purity was (1.8-2.1, The results of DNA extraction and PCR products (280 bp) were shown in Figures 1A and 1B, PCR product were identical with virtual amplifications which used latter in multiple comparison .

The mtDNA sequences show different mutations in study groups in different sites, there were among 173 nucleotides of the D-loop (16034-16207) as showed in Table 1, there were 19 sites differentiated in non-significant differences, about 8 types of substitutions mutation were observed included (A>C, A>G, T>C, G>A, C>T, C>A, C>G, G>A), insertion and deletion mutations also founded, non-significant differences were appeared in the types and number of mutations in this segment of mtDNA as showed in Table 2 and Figure 2.

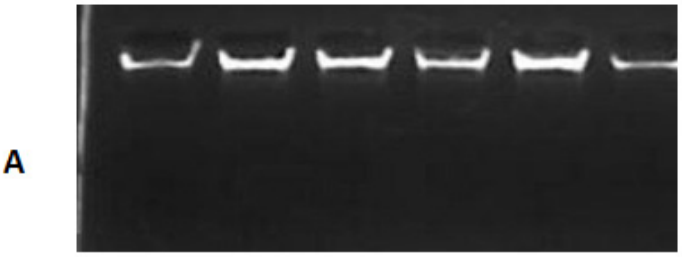

Figure 1A. The electrophoresis of DNA extraction from study subjects and PCR products for all samples.

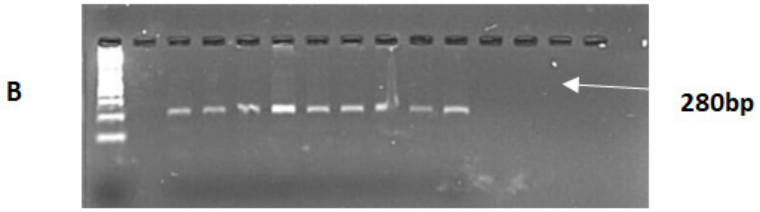

Figure 1B. The electrophoresis of DNA extraction from study subjects it has 280 bp of HV1a 1agarose, 75 V, 20 mA for 30 minutes.

Table 1. The mtDNA mutation loci in depression patients and control.

mtDNA loci	NCBI	Patients (%)	Control	Odd ratio (CI%)	Sig
16034	A	C (10%)	A(100%)	3.3158 (0.1200 to 91.607)	0.4790
16040	A	Deletion (10%) G (10%)	A (100%) A(100%)	3.3158 (0.1200 to 91.607) 3.3158 (0.1200 to 91.607)	0.4790 0.4790
16077	N	T(10%)	A(100%)	3.3158 (0.1200 to 91.607)	0.4790
16129	T	C(30%)	T(100%)	9.8000 (0.4380 to 219.259)	0.1500
16148	G	A(10%)	A(10%)	1.0000 (0.0538 to 18.574)	1
16156	G	A(10%)	G(100%)	3.3158 (0.1200 to 91.607)	0.4790
16166	A	G(10%)	A(100%)	3.3158 (0.1200 to 91.607)	0.4790
16172	C	C	T(10%)	0.3016 (0.0109 to 8.3321)	0.4790
16175	T	T	C(10%)	0.3016 (0.0109 to 8.3321)	0.4790

16186	A	C(10%)	C(10%)	9.0000 (0.8088 to 100.143)	0.0739
16187	-	-	Insertion	133.00 (4.8140 to 3674.45)	0.0039*
16188	C	T(10%)	C(100%)	3.3158 (0.1200 to 91.607)	0.4790
16191	C	T(10%)	C(100%)	3.3158 (0.1200 to 91.607)	0.4790
16193	A	C(10%)	C(50%)	0.1111 (0.0100 to 1.236)	0.0739
16201	C	G(10%)	G(50%)	0.1111 (0.0100 to 1.236)	0.0739
16204	A	T(20%)	A(100%)	6.1765 (0.2599 to 146.78)	0.2600
16205	C	C	A(50%)	0.0476 0.0022 to 1.0293	0.0522
16206`	A	C(10%)	A(100%)	3.3158 (0.1200 to 91.607)	0.4790
16207	G	G	A(50%)	0.0476 0.0022 to 1.0293	0.0522

Note: *Significance< 0.005

Table 2. The mutation type frequency in depression patients and control.

Mutation types	P	C	Odd ratio (CI 95%)	Sig 0.005
A>C	3	2	1.7143 (0.2192 to 13.407)	0.607
A>G	2	0	6.1765 (0.2599 to 146.785)	0.2600
T>C	1	0	3.3158 (0.1200 to 91.607)	0.479
G>A	2	1	2.2500 (0.1701 to 29.768)	0.5383
C>T	2	1	2.2500 (0.1701 to 29.768)	0.5383
C>A	0	1	0.3016 (0.0109 to 8.332)	0.4790
C>G	1	1	1 (0.0546 to 18.30)	1
G>A	2	2	1 (0.1118 to 8.9473)1	1
Deletion	2	0	6.1765 (0.2599 to 146.785	0.2600
Insertion	0	1	0.3016 (0.0109 to 8.332)	0.4790
Total	15	9	1.7300 (0.7359 to 4.0670)	0.2088
	173	173		

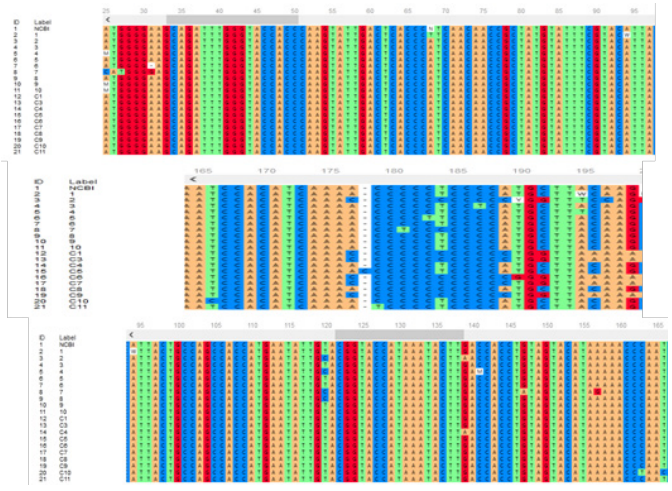

Figure 2. The D-loop mutation in depression patients and control group.

Discussion

Mitochondria have bioenergetics role in cells, furthermore any mutation or DNA damage in DNA can be led to some disease, and abnormalities in mitochondrial DNA have a key role in neurodegenerative disorders [13]. Interestingly, the accumulation of mitochondrial nucleotide changes and the instability of mtDNA have been linked to a variety of illnesses, including cancer and neurological disorders. The D-loop area is crucial in mitochondrial genome replication, transcription, and organization, and mutations in this region have been linked to mitochondrial genome instability [14].

The result of this study agreement with a study conducted by Munakata, et al. that explain that there is no associated between depression and mDNA mutation and this study explain that depression are caused by mutation in mDNA , in contrast with current study [15], several studies showed that Patients with MDD exhibited a greater mtDNA copy number than control individuals, and these findings were consistent whether the patient experienced a single episode or recurring MDD [4]. The reasons for the inconsistencies in previous findings of mtDNA copy number of patients with depression remain unclear. Some studies have reported higher mtDNA copy numbers in MDD cohorts [16,17], while others have reported lower mtDNA copy numbers in individuals with depression, or no changes in mtDNA copy number [18,19].

The discrepancies might be attributed to variation in individuals' medication status, disease duration, study population age compositions (young vs. elderly), challenging to homogenize phenotypes of diverse MDD diagnoses, and/or comorbidity of somatic disorders [20,21].

As a result of Reactive Oxygen Species (ROS) elevation level, which production in the organelle and the low level of Mismatch Repair (MMR) the DNA of mitochondria is highly susceptible to mutations, Although mutations observed throughout the mitochondrial genome, the D-loop is the most variable part of the human mitochondrial genome in addition to other mutations in different sites of genome, So we need to more study and whole mtDNA sequencing to more demonstrations of variations associated with major depression [22-24].

Conclusion

The current study found that the depression disorder didn't relate to HV1 variation in compare with control group, So we need more studies and whole mtDNA sequencing to more demonstrations of variations associated with major depression disorder. The Mitochondria organelle is important for energy production, lipid and steroid metabolism, and cellular stability, as well as modulating Ca^{2+} levels, maintaining ROS levels, and controlling apoptosis.

References

1. Scheffler, Immo E. Mitochondria. New Jersey: Wiley, USA, (2011).
2. Rezin, Gislaine T., Cinara L. Gonçalves, Juliana F. Daufenbach and Daiane B. Fraga, et al. "Acute Administration of Ketamine Reverses the Inhibition of Mitochondrial Respiratory Chain Induced by Chronic Mild Stress." *Brain Res Bull* 79 (2009): 418-21.
3. Sheline, Yvette I., Mokhtar H. Gado and Helena C. Kraemer. "Untreated Depression and Hippocampal Volume Loss." *Am J Psychiatry* 160 (2003): 1516-8.
4. Chung, Jae Kyung, Soo Young Lee, Mira Park and Eun-Jeong Joo, et al. "Investigation of Mitochondrial DNA Copy Number in Patients with Major Depressive Disorder." *Psychiatry Res* 282 (2019): 112616.
5. Manji, Husseini, Tadafumi Kato, Nicholas A. Di Prospero and Seth Ness, et al. "Impaired Mitochondrial Function in Psychiatric Disorders." *Nat Rev Neurosci* 13 (2012): 293-307.
6. Schloesser, Robert J., Husseini K. Manji and Keri Martinowich. "Suppression of Adult Neurogenesis Leads to an Increased HPA Axis Response." *Neuroreport* 20 (2009): 553.
7. Jou, Shaw-Hwa, Nan-Yin Chiu and Chin-San Liu. "Mitochondrial Dysfunction and Psychiatric Disorders." *Chang Gung Med* J 32 (2009): 370-9.
8. Schapira, Anthony HV. "Mitochondrial Disease." *Lancet* 368 (2006): 70-82.
9. Inczedy-Farkas, Gabriella, Viktoria Remenyi, Aniko Gal and Zsofia Varga, et al. "Psychiatric Symptoms of Patients with Primary Mitochondrial DNA Disorders." *Behav Brain Funct* 8 (2012): 1-9.
10. Mancuso, Michelangelo, Daniele Orsucci, Elena Caldarazzo Ienco and Eleonora Pini, et al. "Psychiatric Involvement in Adult Patients with Mitochondrial Disease." *Neurol Sci* 34 (2013): 71-74.
11. Montier, Laura L. Clay, Janice J. Deng and Yidong Bai. "Number Matters: Control of Mammalian Mitochondrial DNA Copy Number." *J Genet Genomics* 36 (2009): 125-31.
12. Yoo, Hee Jeong, Mira Park and Soon Ae Kim. "Difference in Mitochondrial DNA Copy Number in Peripheral Blood Cells between Probands with Autism Spectrum Disorders and their Unaffected Siblings." *World J Biol Psychiatry* 18 (2017): 151-6.
13. Mancuso, Michelangelo, Fabio Coppede, Lucia Migliore and Gabriele Siciliano, et al. "Mitochondrial Dysfunction, Oxidative Stress and Neurodegeneration." *J Alzheimers Dis* 10 (2006): 59-73.
14. Ferreira, Ildete L., Maria V. Nascimento, Márcio Ribeiro and Sandra Almeida, et al. "Mitochondrial-Dependent Apoptosis in Huntington's Disease Human Cybrids." *Exp Neurol* 222 (2010): 243-55.
15. Munakata, Kae, Kumiko Fujii, Shinichiro Nanko and Hiroshi Kunugi, et al. "Sequence and Functional Analyses of mtDNA in a Maternally Inherited Family with Bipolar Disorder and Depression." *Mutat Res* 617 (2007): 119-24.
16. Nicod, Jérôme, Stefanie Wagner, Frederick Vonberg and Amarjit Bhomra, et al. "The Amount of Mitochondrial DNA in Blood Reflects the Course of a Depressive Episode." *Biol Psychiatry* 80 (2016): e41-e42.
17. Tyrka, Audrey R., Stephanie H. Parade, Lawrence H. Price and Hung-Teh Kao, et al. "Alterations of Mitochondrial DNA Copy Number and Telomere Length with Early Adversity and Psychopathology." *Biol Psychiatry* 79 (2016): 78-86.
18. Chang, Cheng-Chen, Shaw-Hwa Jou, Ta-Tsung Lin and Te-Jen Lai, et al. "Mitochondria DNA Change and Oxidative Damage in Clinically Stable Patients with Major Depressive Disorder." *PLoS One* 10 (2015): e0125855.
19. Kim, Moo-Young, Ji-Won Lee, Hee-Cheol Kang and Eosu Kim, et al. "Leukocyte Mitochondrial DNA (mtDNA) Content is Associated with Depression in Old Women." *Arch Gerontol Geriatr* 53 (2011): e218-e221.
20. He, Ying, Jinsong Tang, Zongchang Li and Hong Li, et al. "Leukocyte Mitochondrial DNA Copy Number in Blood is not associated with Major Depressive Disorder in Young Adults." *PLoS One* 9 (2014): e96869.
21. Verhoeven, J. E., D. Révész, M. Picard and E. E. Epel, et al. "Depression, Telomeres and Mitochondrial DNA: Between-and within-Person associations from a 10-Year Longitudinal Study." *Mol Psychiatry* 234 (2018): 850-7.
22. Levy, Richard J. and Clifford S. Deutschman. "Deficient Mitochondrial Biogenesis in Critical Illness: Cause, Effect, or Epiphenomenon?" *Crit Care* 11 (2007): 1-2.'
23. Kadhum, Aamal Muhsen, Bayader M. Abd Al-Kadim, Thanaa Chasib Kareem and Ali Ahmed Nayyef, et al. "Associations of Tumor Necrosis Factor-[alpha] G-308A (rs1800629) Gene Polymorphisms with Major Depressive Disorder." *Neuro Quantology* 19 (2021): 133-7.
24. Kadhum, Aamal Muhsen, Eman Salih Abd, Ruaa Amer Taleb and Kareem Nasir Hussain, et al. "Genetic Polymorphisms of Mitochondrial Genes (ND1, ATP6c) in Depression Patients." *Neuro Quantology* 19 (2021): 128-33.

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