To the results of the study of the association of ALA54THR polymorphism of the FABP2 gene in patients with vitiligo of the Uzbek population of the Bukhara region

Shaxnoza Zakirovna Mavlyanova
Republican Specialized Scientific and Practical Medical Center for Dermatology and Venereology of the Ministry of Health of the Republic of Uzbekistan, Tashkent, Uzbekistan
shahnoza_m@mail.ru
Ixlosjon Boltayevich Shukurov
Republican Specialized Scientific and Practical Medical Center for Dermatology and Venereology of the Ministry of Health of the Republic of Uzbekistan, Tashkent, Uzbekistan
ihlosboltaev@gmail.com.
Malohat Farmonovna Yaxshiyeva
Bukhara State Medical Institute, Bukhara, Uzbekistan
malohatyaxshiyeva@gmail.com

ABSTRACT
The aim of our research is to study the allelic variants and association genotypes of the Ala54Thr polymorphism of the FABP2 gene in patients with vitiligo and to identify predictors of the development of the disease. Material and methods of research: The object and subject of the study were patients with vitiligo, DNA samples of patients and healthy donors, the Ala54Thr gene of the FABP2 gene. The study included 109 patients with vitiligo aged 5 to 62 years old, who were observed at the clinic of the RSNPMC DVIC of the Ministry of Health of the Republic of Uzbekistan and "Ixlos Med service" LLC. Conclusions. Allele Thr and heterozygous genotype of A la /T hr polymorphism gene FABP 2 are markers of an increased risk which results in developing vitiligo, especially localized and generalized forms of dermatosis, in the Uzbek population in the Bukhara region.
KEYWORDS*:* vitiligo, genetics, ALA54THR of the FABP2 gene, clinic, prognosis.

INTRODUCTION

The article presents the results of molecular genetic studies of the ALA54THR gene FABP2 in patients with vitiligo. 109 patients with vitiligo aged from 5 to 62 years were examined. All patients have underwent clinical, molecular, genetic and statistical studies. The results of the study showed that the allele Thr and heterozygous genotype of A la /T hr polymorphism gene FABP 2 are markers of an increased risk of developing vitiligo in the Uzbek population of the Bukhara region, especially the localized and generalized form of dermatosis. ($\chi 2$ =15 p< 0.0001; OR =5.67; 95% CI 2.14-14.7; $\chi 2$ = 11.6 p<0.003; OR =3.21; 95% CI 1.14 -9.0). Allele A la and functionally favorable genotype of **A** la / Ala are reliable protective markers in relation to the development of pathology (χ^2 =11.6 p<0.003; OR =0.21; 95% CI 0.08-0.57).

Genetically determined individual differences in metabolic disorders of the body in many chronic diseases, including skin pathology, determine the relevance of studying polymorphisms (structural changes) of genes that regulate metabolism and human eating behavior [1,2,5-11].

It should be noted that despite the study of the pathogenesis of vitiligo by experimental and clinical studies, there is still no evidence-based theory about the presence of specific autosensitization to melanocytes, the main metabolic processes that play an important role in melanogenesis, as well as in the progression of depigmentation.

Up to now, many single nucleotide polymorphisms (SNPs) of genes associated with the regulation of lipid and carbohydrate metabolism, adipogenesis, thermoregulation, circadian rhythm and eating behavior are known [12, 15]. Thus, the Ala54Thr (rs1799883 G/A) polymorphism is one of the most studied polymorphisms of the gene encoding a protein that binds type 2 fatty acids (fatty acid binding protein 2 - FABP2). It has been established that the FABP2 protein is involved in the intracellular transport and metabolism of long-chain fatty acids, and it also takes part in several stages of absorption and transport of lipids in the intestine [4, 9].

The aim of our research is to study the allelic variants and association genotypes of the Ala54Thr polymorphism of the FABP2 gene in patients with vitiligo and to identify predictors of the development of the disease.

MATERIALS AND METHODS

The object and subject of the study were patients with vitiligo, DNA samples of patients and healthy donors, **the Ala54Thr** gene of the FABP2 gene. The study included 109 patients with vitiligo aged 5 to 62 years old, who were observed at the clinic of the RSNPMC DVIC of the Ministry of Health of the Republic of Uzbekistan and “Ixlos Med service” LLC. Among 109 patients, there were 52 females and 57 males. The diagnosis in all patients was confirmed by the results of a clinical examination (dermatoscopy). The control group consisted of 81 healthy individuals without skin pathology of the corresponding age.

D classification. Mosher (1979) diagnosed 40 patients with a localized form, which accounted for 36.7%, and 46 patients with a generalized form, which accounted for 42.2% of cases. The universal form was diagnosed in 23 out of 109, which accounted for 21.1% of cases, respectively. In the group of patients with a localized form among 40 patients, 15 (37.5%) of them noted the focal form and the other 25 (62.5%) the segmental form of vitiligo. Among 46 patients with a generalized form, 11 (23.9%) were diagnosed with a vulgar form and 35 with an acrofacial form, which accounted for 76.1% of cases.

Molecular genetic examination of biomaterials (DNA) was performed on the basis of “GENOTEXNOLOGIYA” LLC according to scientific agreement. The object and subject of the study were DNA samples of sick and healthy individuals, gene Ala54Thr of the FABP2 gene**.**

DNA samples were isolated from peripheral blood lymphocytes according to a modified method. The concentration and purity of the isolated DNA were evaluated by measuring the optical density of DNA-containing solutions at a wavelength of 260 and 280 nm against TE on a NanoDrop 2000 spectrophotometer (USA). Genotyping of the Ala54Thr polymorphism of the FABP 2 gene was performed on a real-time PCR amplifier Rotor Gene 6000 Model 65H0-100 (Australia), using the test system of the company " Synthol " Cat. No.-NP_555_100_RG (Russia), according to the manufacturer's instructions. Statistical analysis of the results was carried out using the statistical software package " OpenEpi 2009, Version 2.3". The frequency of variants of alleles and genotypes

(f) was calculated by the formula: f = n /2 N and f = n / N , where n is the occurrence of the variant (allele and genotype), N is the sample size.

Statistical analysis of the results was carried out using the statistical software package " OpenEpi 2009, Version 2.3".

RESULTS AND DISCUSSION

Results of molecular genetic studies of the Ala 54 Thr gene FABP 2 (rs1799883 G/A) are presented in the table

Table 1. Frequency distribution of alleles and genotypes of Ala 54 Thr gene polymorphism FABP 2 (rs1799883 G/A) in groups of patients and control n is the number of examined patients; * n - number of studied alleles

No.	**Group**	**Allele frequency**				**Frequency distribution of genotypes**					
		Ala		Thr		**A** la / Ala		**A** la / **T** hr		**T** hr /**T** hr	
		* n	%	* n	%	n	%	n	%	n	%
1.	Main group n =109 (218)	185	84.8	33	15.1	83	76.1	19	17.4	7	6.4
2	Counter group n =81 (162)	157	96.9	5	3.1	76	93.8	5	6.2		

Our study of the distribution frequency of polymorphism alleles Ala 54 Thr of the FABP 2 gene in the sample of patients of the main group and in the population sample (control) showed that the mutant allele "T hr " was significantly more common in patients with vitiligo than in the control (15.1% and 5%, respectively; (χ^2 =15; p <0.0001; OR =5.6; 95% CI 2.14-14.7); Whereas, the frequency of the "wild" allele "A la " in patients with vitiligo was detected in 84.8% of cases (185/218) , and in the control group it was 96.9% (157/162), which is 1.1 times higher than in the main group (χ^2 =15; p< 0.0001; OR =0.18; 95% CI 0 .07-0.47).

Comparative assessment of the frequency of occurrence of the mutant variant of the T hr allele among patients with vitiligo (15.1%) and in controls (5%) showed a significant difference in the values of the studied parameter (χ^2 =15; p<0.0001; OR =5.6;95% CI 2.14-14.7). The result obtained may indicate the relationship of the allele "T hr " polymorphism Ala 54 Thr gene FABP 2, leading to the replacement of alanine by tyrosine at position 54 of the amino acid sequence, with the development of a violation of the melanogenesis process. At the same time, the risk of developing vitiligo in the presence of a variant T hr in the genome polymorphism allele increased by 5.6 times (OR = 5.6).

Analysis of the distribution of genotypic variants of the rs1799883 G/ A (Ala 54 Thr) polymorphism of the FABP 2 gene revealed the predominance of the heterozygous variant **A** la **/T** hr 17.4% (19/109) in patients with vitiligo, while in the group of control healthy individuals this genotype **A** la **/T** hr was 6.2% (5/81), respectively, which was 2.8 times lower compared with the main group of patients with vitiligo. (χ^2 =11.6; p< 0.003; OR =3.2 95% CI 1.14-9.0).

Mutant homozygous **T** hr **/T** hr FABP 2 gene in the group of patients with vitiligo was 6.4% (7/109), and this genotype was not detected in the group of control individuals. (χ^2 =11.6; p< 0.003; OR =11.93 95% CI 0.67-211.9). At the same time, the functional genotype **A** la **/** Ala gene FABP 2 in the control group was determined by 93.8% (76/81), while in the group of patients with vitiligo

genotype **A** la / Ala amounted to 76.1% of cases (83/109), which was 1.2 times lower compared to the control group. (χ^2 =11.6; p< 0.003; OR =0.21 95% CI 0.08-0.6)

The obtained data may indicate that the heterozygous genotype **A** la /**T** hr of the rs1799883 G/ A (Ala 54 Thr) polymorphism of the FABP 2 gene is a genetic determinant that determines the formation of a violation of melanogenesis , and its carriage is a predisposition factor for the development of vitiligo, increasing its risk by 3.2 times (OR = 3.21).

Comparison with the clinical course of vitiligo with associations of genotype polymorphisms of the Ala 54 Thr gene of the FABP 2 gene revealed the following features (Table 2).

Table 2. Frequency distribution of alleles and genotypes of the rs1799883 G/ A (Ala 54 Thr) polymorphism of the FABP 2 gene in patient groups is the number of examined patients; *n - number of studied alleles

Group	**n**	**Allele frequency**				**Frequency distribution of** FABP 2 genotypes					
		Ala		Thr		**A** la / Ala		**A** la / **T** hr		**T** hr /**T** hr	
		n	%	n	%	n	%	N	%	n	%
Localized form of vitiligo	40 (80)	67	83.8	13	16.3	31	77.5	5	12.5	4	10
Generalized form of vitiligo	46 (92)	74	80.4	18	19.6	31	67.4	12	26.1	3	6.5
Universal form	23 (46)	44	95.6	2	4.3	21	91.3	2	8.6	-	-
Control group	81 (162)	157	96.9	5	3.1	76	93.8	5	6.2		

So, taking into account the clinical form of vitiligo in the localized form, the frequency of heterozygous **A** la /**T** hr and mutant **T** hr /**T** hr genotypes of the FABP 2 gene occurred in 12.5% and 10% of cases (5/40; 4/40), which is 2 .02 times higher than the indicators of control persons (χ^2 =10.2; p < 0.006; OR-2.2; 95% CI-0.6-7.9; χ^2 =10.2; p < 0.006; OR- 20.1, 95% CI-1.05-383.1 respectively).

And with a generalized form of vitiligo, the frequency of heterozygous **A** la /**T** hr and mutant **T** hr /**T** hr genotypes of the FABP 2 gene occurred in 26.1% and 6.5% of cases (12/40; 3/40), which was 4.2 times higher than in control individuals (χ^2 =16.3, p < 0.00005, OR-5.36, 95% CI-1.7-16.4, $\chi 2^{=}$ 16.3, p < 0.00005, OR-13.1, 95% CI-0.6-259.7, respectively). The data obtained were statistically significant. (P <0.05). Whereas in the universal form, the frequency of heterozygous genotypes **A** la /**T** hr genotypes of the FABP 2 gene occurred in 8.6% of cases (2/46) (χ^2 =0.18; p<0.9 OR =1.45 95% CI 0.26-8.0) and was not statistically significant. (P >0.05).

Analysis of the data obtained indicates that the mutant Thr allele and the heterozygous **A** la /**T** hr variant of the FABP 2 gene is a significant risk criterion for impaired melanogenesis and the development of vitiligo, especially a localized and generalized form of the disease) Fig. 1)

We have analyzed the expected and observed frequencies of the genotypes of the studied polymorphisms and the correspondence of the frequency distribution to the *Hardy - Weinberg equilibrium (HV).* (table 3*).*

Table 3. Expected and observed frequency of distribution of genotypes according to RHB of polymorphism rs1799883 (Ala 54 Thr) of FABP 2 gene in the main group of patients with vitiligo

Genotypes	genotype frequency		χ^2	R
	observed	expected		
A la / Ala	76.2	53, 0 1	0.720 _ _	
A la / **T** hr	17,4	39.6 _	0.257 _ _ _	
T hr /**T** hr	6.42	7.4 _	0.023	
Total	100.00	100.00	4.65	**0.03**

As follows from Table 3, the frequency distribution of genotypes for RHV polymorphism rs1799883 (Ala 54 Thr) of the FABP 2 gene in the main group of patients with vitiligo, the observed frequency of functional homozygous genotypes A la / Ala occurred in 76.2%, and the expected one was 53.01% of cases. While the observed heterozygous genotype A la /T hr was 17.4%, and the expected heterozygous genotype was 39.6 cases, which was 2.3 times higher than the observed indicators. Homozygous unfavorable variant of genotypes - T hr /T hr gene FABP 2 in the observed frequencies was 6.4%, and in the expected frequencies it was 7.4%, which increased by 1.2 compared to the observed values, respectively. The results obtained are of great importance in predicting the risk of developing morbidity.

Whereas in the control group, the observed and expected frequency of favorable genotypes **A** la / Ala of the FABP 2 gene was 93.8% and 60.6%, respectively, and the observed frequency of the heterozygous variant **A** la /**T** hr was 6.17% and the expected frequency was 34.5%, respectively, which was 5.6 times higher than observed frequencies. ($P < 0.05$).

Whereas the homozyotic variant of favorable genotypes **T** hr /**T** hr the observed frequency was - 0, and the expected frequency was 4.9%, respectively (Table 4).

Table 4. Expected and observed frequency of distribution of genotypes according to RHB of polymorphism rs1799883 (Ala 54 Thr) of FABP 2 gene in the control group of healthy individuals.

Genotypes	genotype frequency		χ^2	R
	observed	expected		
A la / Ala	93.8	60.6 _	0.939	
A la / **T** hr	6.17	34.5 _	0.060	
T hr /**T** hr	0	4.9	0.001	
Total	100.00	100.00	0	**1**

Comparative analysis of the expected and observed genotype frequencies of the rs1799883 (Ala 54 Thr) polymorphism of the FABP 2 gene showed a statistically significant deviation of indicators ($P<0.05$) in all studied groups and subgroups. This fact indicates that the observed proportion of genotypes in the studied samples corresponds to the Hardy- Weinberg equilibrium.

As we can understand, the allele Thr and heterozygous genotype of A la /T hr polymorphism gene FABP 2 are markers of an increased risk of developing vitiligo in the Uzbek population of the Bukhara region, especially in localized and generalized forms of dermatosis. (χ^2 =15 p< 0.0001; OR =5.67; 95% CI 2.14-14.7; $\chi 2^{=}$ 11.6 p<0.003; OR =3.21; 95% ^{CI} 1.14 -9.0). Allele A la and functionally favorable genotype **A** la / Ala are reliable protective markers in relation to the development of pathology (χ^2 =11.6 p<0.003; OR =0.21; 95% CI 0.08-0.57).

CONCLUSION

Allele Thr and heterozygous genotype of A la /T hr polymorphism gene FABP 2 are markers of an increased risk which results in developing vitiligo, especially localized and generalized forms of dermatosis, in the Uzbek population in the Bukhara region. ($\chi 2$ =15 p< 0.0001; OR =5.67; 95% CI 2.14-14.7; $\chi 2$ = 11.6 p<0.003; OR =3.21; 95% CI 1.14 -9.0). Allele A la and functionally favorable genotype **A** la **/** Ala are reliable protective markers in relation to the development of pathology (χ^2 =11.6 p<0.003; OR =0.21; 95% CI 0.08-0.57).

REFERENCES

1. Abduraxmanov, D.S., Kurbaniyazov, Z.B., Davlatov, S.S., & Rakhmanov, K.E. (2020). Criteria for choosing surgical treatment of patients with ventral hernias and obesity// International Journal of Pharmaceutical Research, 12, – P. 4057–4066.
2. Aminov Z. Z. Khakimova S. Z. Davlatov S. S. (2020). Improvement Of Treatment Protocols Of Pain Syndrome In Patients With Chronic Brucellosis. European Journal of Molecular & Clinical Medicine, 7(3), 2540-2545.
3. Bozorov, A.G., Ikhtiyarova, G.A., & Davlatov, S.S. (2020). Biochemical markers for prediction of premature labor in urogenital infections // International Journal of Pharmaceutical Research, 12, – P. 4894–4897.
4. Davlatov, S., Rakhmanov K., Qurbonov N., Vafayeva I., & Abduraxmanov D. (2020). Current State of The Problem Treatment of Mirizzi Syndrome (Literature Review)// International Journal of Pharmaceutical Research, 12, – P. 1931-1939. DOI:https://doi.org/10.31838/ijpr/2020.SP2.340
5. Ikhtiyarova, G. A., Dustova, N. K., Khasanova M. A., Suleymanova G. S., & Davlatov, S. S. (2021). Pathomorphological changes of the placenta in pregnant women infected with coronavirus COVID-19. International Journal of Pharmaceutical Research, 13(1), 1935-1942. doi: 10.31838/ijpr/2021.13.01.283
6. Kudratova, D. Sh, Ikhtiyarova, G.A., & Davlatov, S.S. (2021). Medical and social problems of the development of congenital malformations during a pandemic. International Journal of Pharmaceutical Research, 13(1), 756-760. doi: 10.31838/ijpr/2021.13.01.130
7. Mavlyanova Sh. Z., Shukurov I. B., Yahshiyeva M. F. //Assessment of the Dermatological Index of the Symptom Scale in Patients with Seborrheic Der-matitis and Opportunistic Infection Based On the Use of Natural Silicon Oxide //Annals of R.S.C.B., Vol. 25, Issue 4, 2021, Pages. 3922 – 3930 Received 05 March 2021; Accepted 01 April 2021.
8. Nasibulina E.S., Borisova A.V., Akhmetov I.I. Study of the association of the Ala54Thr polymorphism of the FABP2 gene with the risk of developing obesity, body fat mass and physical activity.//Question of nutrition. – 2013- №5-p. 23-28.
9. Oripov, F., Blinova, S., Dekhkanov, T., & Davlatov, S. (2020). Development of immune structures of the leaning intestine of rabbits in early postnatal ontogenesis. International Journal of Pharmaceutical Research, 13(1), 299-301. doi:10.31838/ijpr/2021.13.01.042
10. Saatov B.T., Karimov Kh.Ya., Arifov S.S., Saatov T.S. Ibragimov Z.Z., Boboev K.T. Study of the relationship of polymorphism 308 G / A of the TNF-α gene with the risk of developing vitiligo // Scientific and practical journal " Dermatovenereology and aesthetic medicine", Tashkent, 2016. - No. 2.- P.75-82.

11. Sabirov U., Azimova F., Toirov B. Muminova S. Evaluation of the effectiveness of surgical treatment of vitiligo by melanocytic transplantation based on the study of TRP-2 molecule expression. // International Journal of Pharmaceutical Research. – Salem, 2021 No. 13(1). SJR=0.28

12. Sabirov U.Yu., Bobobekov K.T., Toshpulatov Sh.Sh. Molecular genetic and immunological parameters of the organism in the study of the clinical efficacy of the complex treatment of vitiligo. // Dermatovenereology and aesthetic medicine: Scientific and practical journal - Tashkent, 2021. - No. 4.p.122-128.

13. Sabirov U.Yu., Toshpulatov Sh.Sh. To determine the possible relationship of polymorphic of TNF-α and TGF- β genes wiyh the risk of developing vitiligo.// Dermatovenereology and aesthetic medicine: Scientific and practical journal - Tashkent, 2021. No. 3 - p.6-7.

14. Shukurov I.B., Yakhshieva M.F., The study of the prevalence and medical and social aspects of vitiligo in the Bukhara region Universum: medicine and pharmacology.Moscow 2021.P. 8-13.

15. Takakura Y., Yoshioka K., Umekawa T. et al. // Diabetes Res. Clinic. Pract. - 2005. - Vol. 67, No. 1. - P. 36-42.

16. Zhao T., Zhao J., Lv J. et al. // Nutr. metab. Cardiovasc. Dis. - 2011. - Vol. 21, No. 10. - P. 823-829.