Obesity Medicine 15 (2019) 100128

Contents lists available at ScienceDirect

Obesity Medicine

journal homepage: www.elsevier.com/locate/obmed

Original research

Associations of polymorphisms of the FTO, ADRB3, LEPR genes with obesity and the impact on Them of a complex of recreational activities among residents of the North Caucasus

Sergey Petrovich Lysenkov[a], Rustem Askarbievich Thakushinov[a], Tamara Ur'evna Urakova[a], Dmitriy Vitalevich Muzhenya[a,*], Aminat Ramazanovna Tuguz[b]

[a] FSBEI HE[a], "Maikop State Technological University", Medical Institute, 385000, Republic of Adygeya, Maikop, Russia
[b] FSBEI HE[a], "Adyghe State University", Republic of Adygeya, Maikop, Russia

ARTICLE INFO

Keywords:
Obesity
Single nucleotide polymorphisms
Hypocaloric diet
Health programs
Molecular genetic markers

ANNOTATION

Objective: To study the effect of a comprehensive health program on the effectiveness of reducing overweight depending on the carriage of allelic variants and FTO (rs9939609), ADRB3 (rs4994) and LepR (rs1137101) genotypes among residents of the North Caucasus.

Materials and methods: Gene polymorphisms were analyzed by the SNP-method with allele-specific primers on the test systems of 'Litex' Research and Production Company (RPC). The frequencies rs9939609, rs4994 and rs1137101 were studied in individuals with overweight, I-III degrees of obesity (n = 120) compared with the control group (n = 42). Experimental data were processed in the program SPSS Statistics 22.0. A comprehensive wellness program (CWP), along with health improving measures, limits caloric intake to 800–1200 kcal per day and excludes animal fats and proteins. The body mass index (BMI) of the examined individuals was calculated by the Ketle method as the ratio of body weight (kg) to height (m^2). In accordance with WHO criteria (1997), the severity of overweight varies in the ranges of values: normal weight – 18–24.9 kg/m^2; overweight – 25–29.9 kg/m^2; I degree obesity – 30–34.9 kg/m^2; II degree obesity – 35–39.9 kg/m^2; III degree obesity – 40 and more kg/m^2.

Results: A significant increase in rs9939609 (p = 0.003), rs4994 (p = 0.0001) was found in the group of patients when compared with the control. The 64Arg allele (p = 5.0E-6) and the Arg64Arg genotype (p = 0.0001) of the ADRB3 gene have the greatest prognostic significance for the diagnosis of obesity, increasing the risk by 10 and 17 times, respectively. Significant differences in the distribution of FTO gene polymorphism were established to a lesser extent and only by the T23525T genotype (χ^2 = 7.20; p = 0.03). However, the correlation analysis revealed a number of strong positive connections for the T23525T FTO genotype between the ratio of visceral fat (r = +0.541; p = 0.01), fat (r = +0.548; p = 0.01) and muscle mass (r = +0.587; p = 0.01). It should be noted that with SNP-typing of three polymorphic variants of the FTO, ADRB3 and LepR genes, the transversion phenomenon was established by rs9939609 in 26 out of 120 participants (21.6%).

Conclusion: According to the obtained data, we confirmed the promising use of rs9939609, rs4994 as genetic markers of obesity. The discovered phenomenon of transversion of polymorphisms in the FTO gene under the influence of the complex health program does not exclude a multi-level regulation of reparation processes and epigenetic activation of genes, which requires further study.

1. Introduction

Obesity is one of the global problems of the 21st century that threatens modern society. According to the final report of 2016 of the World Health Organization (WHO), more than 1.9 billion adults (over 18) and 41 million children under the age of 5 years were overweight and obese in 2015. In total, from 1980 to 2015, this figure more than doubled worldwide (World health organization, 2013). From the point of view of clinical diagnosis, obesity is considered as lipid metabolism disorder, due to the energy imbalance of the amount of the consumed

* Corresponding author. Department of Pathomorphology and Clinical Pathophysiology, Medical Institute of the Maikop State University of Technology, Russia.
E-mail address: dmuzhenya@mail.ru (D.V. Muzhenya).
[a] FSBEI HE – Federal State Budget Educational Institution of Higher Education.

https://doi.org/10.1016/j.obmed.2019.100128
Received 26 May 2019; Received in revised form 7 August 2019; Accepted 7 August 2019

food and energy (World health organization, 2013; Hindorff et al., 2009).

According to the WHO, overweight is one of the main risk factors for the development of type 2 diabetes, endocrine disorders, cardiovascular, oncological and other noncommunicable diseases, the annual loss of which is about 36 million people (63% of cases). More than 14 million people die prematurely (before the age of 50); and, while maintaining the current trends by 2030, these problems, taking on the character of pandemics, can kill 52 million people each year according to forecasts of WHO experts (World health organization, 2013). According to S.I. Kuzin, M.V. Karmanov (2016), about 40% of the working-age population in Russia are overweight and have obesity of varying degrees (Hindorff et al., 2009; Kuzin and Karmanov, 2016).

Taking into account the achievements of modern molecular biology, identification of causal relationships in the development of obesity and socially significant diseases will contribute to the promotion of health and the prevention of noncommunicable diseases; prevent depopulation processes in the whole world including Russia. However, the concept of the development of obesity under the influence of risk factors, established by WHO, does not provide exhaustive ideas about the trigger mechanisms for the occurrence of pathologies and the role of heredity.

According to the "HuGE Literature Finder" and the "National Center for Biotechnology Information" polymorphisms of the genes, involved into the pathogenesis of obesity "Fat mass and obesity-associated protein" (FTO), adrenoreceptor 3-type β (ADRB3) and leptin receptor (LEPR) regulating lipid balance and energy processes, have a significant impact on the proteome (HuGE Literature Finder, 2019; The National Center for Biotechnology Information, 2019).

A large number of single nucleotide polymorphisms (SNP – single nucleotide polymorphism) with different prognostic significance (The Allele Frequency Database, 2019; Puzyrev et al., 2007) was revealed during gene sequencing. However, according to the international project "1000 GenomesProject", the variation in the frequency of polymorphisms indicates the unequal degree of their association with obesity for different ethnic groups. Therefore, conducting regional studies to analyze the distribution and association of SNP with the risk of developing metabolic syndrome is necessary. Despite the identified positive correlation of gene polymorphisms with overweight and other functional parameters, the combined effect of mutations, associated with obesity and modeling of eating behavior and exercise on body weight correction, has not been studied.

The prospect of studying the effect of diet and exercise on the effectiveness of reducing body mass index, depending on the carriage of allelic variants and genotypes, will allow to identify molecular-genetic mechanisms affecting susceptibility to obesity, as well as to identify possible ways of correcting point mutations of the genes responsible for developing obesity.

2. Purpose of the work

A study of the effect of a comprehensive health program on the effectiveness of the reduction of body mass index depending on the carrier of allelic variants and FTO (rs9939609), ADRB3 (rs4994) and LepR (rs1137101) genotypes among residents of the North Caucasus.

2.1. The contingent of surveyed individuals

The control group is represented by unrelated donors (n = 42) aged 20–60 years, without the hereditary burden and clinical manifestations of chronic diseases, which is confirmed by the database of the Adygei Republican Blood Transfusion Station. The second group consisted of (n = 120) patients of the clinic of the Health Center, LLC (Maykop, Republic of Adygeya, Russia) aged 21–63 years with overweight, I-III degrees of obesity and with concomitant diseases (type 2 diabetes, arterial hypertension, chronic pancreatitis, etc.). Both surveyed groups were represented by residents of the North Caucasus region.

Table 1
The list of concomitant diseases in the study group (n = 120 persons).

№	Concomitant diseases	%
1	Hypertension	40,8
2	Coronary heart disease	2,5
3	Atherosclerosis	2,5
4	Type diabetes mellitus	15,8
5	Chronic bronchitis	4,2
6	Bronchial asthma	1,7
7	Thyroid diseases (goiter, hypothyroidism, hyperthyroidism, autoimmune thyroiditis)	14,2
8	Chronic pancreatitis	13,3
9	Chronic gastritis	3,3
10	Gallstone disease	10
11	Urolithiasis	20
12	Arthritis. Arthrosis	29,2
13	Osteochondrosis	44,6

The body mass index (BMI) of the examined individuals was calculated by the Ketle method as the ratio of body weight (kg) to height (m^2). In accordance with WHO criteria (1997), the severity of overweight varies in the ranges of values: normal weight – 18–24.9 kg/m^2; overweight – 25–29.9 kg/m^2; I degree obesity – 30–34.9 kg/m^2; II degree obesity – 35–39.9 kg/m^2; III degree obesity – 40 and more kg/m^2. Concomitant diseases are presented in Table 1.

2.2. Research methods

A comprehensive survey included: measurement of anthropometric data and standard biochemical blood parameters (total cholesterol, high/low lipoproteins, triglycerides). The body mass index (BMI) was calculated using the Quetelet method (IR = mass to body height (kg)/height ratio (m^2)). A quantitative assessment of muscle and fat body mass was performed using the devices "Tanita" (Japan) and DDFAO (France; CE certificate 0459, which allows use as medical equipment in European countries). The device meets the class of medical equipment type BF in accordance with IEC6011/EN60601-1. The principle of operation of the device is based on impedancemetry. By means of six electrodes, placed on the soles of the feet, hands and head, the electrical conductivity of the tissues is measured at various frequencies, specified by the device. Before and after the CWP course, the following was studied: total fat mass (kg), visceral fat (kg), muscle mass (kg), lean mass (kg), total fluid (kg), intracellular water (kg), extracellular water (kg).

As a result of the comprehensive wellness program, the adjustment of the body mass index had been carrying out for 19–21 days. The complex of health procedures included: low-calorie meals (800–1200 kcal/day) excluding animal proteins and fats from the diet of animals, cleansing enemas or colon hydrotherapy (2–3 times a week); laser and bio-resonance therapy, massage, warm shower (2–3 times a day), use of the infrared sauna (1 time per week). Contact laser therapy with a projection on the region of the spleen was carried out 2–3 times a week by an infrared laser with a wavelength of 0.85–0.89 μm using the "Milta" apparatus (ZAO "NPO Cosmic Instrument Making"). The courses of bio-resonance therapy (6–8) were carried out in the mode, specified with an interstitial scanner EIS CE 0535 (ESTECK System Complex, USA).

The genomic DNA of the examined individuals was isolated from whole blood leukocytes using DNA express-blood reagent (NPF "Litekh" (Russia)). The purity of the DNA samples was tested on a NanoDrop 2000c spectrophotometer (Thermo Scientific, USA). Polymorphisms of the FTO genes (A23525T, rs9939609), ADRB3 (Trp64Arg, rs4994) and LepR (Gln223Arg, rs1137101) were studied in DNA samples of donors and patients, which were typed using commercial test-systems of NPF "Litekh" (Russia) with electrophoreteras. on the basis of the

"Immunogenetic Laboratory" of the Research Institute of Complex Problems of the Adyghe State University (Maykop, Russia). In order to verify the obtained data, DNA samples were SNP-typed in the independent laboratory "Development Biology and Genome Organizations" (Rostov-on-Don, Russia) with the obligatory condition of observing identical experimental conditions.

Genotype distributions corresponded to expected values at Hardy-Weinberg equilibrium and compared frequencies of allelic variants/genotypes using χ^2 (chi-square) for contingency tables 2x2 corrected by Iates for continuity and calculating the odds ratio (odds-ration or OR), 95% confidence interval (95% CI). The non-parametric Spearman method in the SPSS Statistics 22.0 program was used to calculate correlations.

The study was conducted in accordance with the principles of the Helsinki Declaration with the written informed consent of all participants in the experiment.

3. The results of the study

According to "HuGE Literature Finder" and "National Center for Biotechnology Information" (HuGE Literature Finder, 2019; The National Center for Biotechnology Information, 2019), allelic variants and FTO genotypes (A23525T, rs9939609), ADRB3 (Trp64Arg, rs4994) and LepR (Gln223Arg, rs1137101) in donor groups (control) and the examined patients (Tables 2 and 3) were typed at the first stage to compare the prognostic significance of the polymorphisms of three genes with impaired metabolic processes and obesity.

According to the obtained results (Table 2), the frequencies rs4994 (ADRB3), rs9939609 (FTO) were statistically significantly ($p < 0.05$) increased in the patient group compared to the control, but the polymorphism of the ADRB3 gene with a high degree of reliability is most significant, as in the homozygous Arg/Arg genotype ($\chi^2 = 18.31$; $p = 0.0001$) and the 64Arg allele ($\chi^2 = 20.74$; $p = 5.0E^{-6}$). Heterozygous Trp/Arg genotype ADRB3 was not detected in patients, which significantly increases the significance of the obtained results.

Carrying the mutant Arg allele and the homozygous genotype of the ADRB3 gene increases the risk of developing the disease 9.95/16.76 times, respectively (Table 2). Significant differences in the frequency distribution of polymorphism of the FTO gene (rs9939609), which sick and donors have, were identified to a lesser extent and only by the T23525T genotype. Gender differences in the distribution and association of allelic FTO variants (A23525T, rs9939609), ADRB3 (Trp64Arg, rs4994) and LepR (Gln223Arg, rs1137101) with the risk of obesity are presented in Tables 3 and 4.

Association with overweight and obesity in men, as opposed to women, was installed only on rs4994 gene ADRB3. At the same time, the carriage of the 64Arg allele increases the risk of developing pathology by more than 28 times, and the homozygous Arg64Arg genotype increases by 20 compared to donors and twice as compared with women (Tables 3 and 4).

An important aspect of the experimental study is to identify the links of the physiological signs of the examined patients with certain genotypes, as well as a correlation analysis in order to identify prognostic unfavorable genetic variants as targets for subsequent therapy (Table 5).

A number of reliable ($p \ll 0.05$) links (Table 5) were identified in a comparative analysis of the dependence of anthropometric indicators on the rs1137101 and rs9939609 genotypes. Patients with heterozygous Gln/Arg LepR genotypes were distinguished by lower weight, BMI, visceral fat content, waist circumference, which is a more favorable prognostic factor (Table 5). The ratios of anthropometric indices for LepR homozygotes Gln/Gln and Arg/Arg practically did not differ; however, individuals with a mutant homozygous genotype had higher studied parameters.

The most significant deviations from normal donor scores and the effects of the complex health program were noted by the T23525T polymorphism (rs9939609) of the FTO gene. The carriage of the homozygous T23525T variant, compared with other genotypes, reveals itself not only with a significant increase in all prognostically unfavorable anthropometric indicators (Table 5) but also with a smaller

Table 2
The frequency of SNP genes in the control group and the group of patients.

Gene	Genotypes/Alleles	Genotype/Alleles Frequencies in Groups		χ^2	P	OR*	
	Genotypes	Patients (all) (n = 120)	Control (n = 42)			value	95% CI
FTO (rs9939609)	A/A	0.350	0.286	7.20	0.03	1.35	0.62–2.90
	A/T	0.400	0.619			0.41	0.20–0.84
	T/T	0.250	0.095			3.17	1.04–9.61
	Alleles						
	A	0.550	0.595	0.52	0.47	0.83	0.50–1.38
	T	0.450	0.405			1.20	0.73–1.99
	Genotypes	Patients (all) (n = 40)	Control (n = 32)				
ADRB3 (rs4994)	Trp/Trp	0.050	0.219	18.13	0.0001	0.19	0.04–0.98
	Trp/Arg	0.000	0.250			0.04	0.00–0.64
	Arg/Arg	0.950	0.531			16.76	3.44–81.59
	Alleles						
	Trp	0.050	0.344	20.74	$5.0E^{-6}$	0.10	0.03–0.31
	Arg	0.950	0.656			9.95	3.21–30.81
	Genotype	Patients (all) (n = 40)	Control (n = 32)				
LepR (rs1137101)	Gln/Gln	0.325	0.156	3.02	0.22	2.60	0.81–8.30
	Gln/Arg	0.450	0.500			0.82	0.32–2.08
	Arg/Arg	0.225	0.344			0.55	0.20–1.57
	Alleles						
	Gln	0.550	0.406	2.94	0.09	1.79	0.92–3.48
	Arg	0.450	0.594			0.56	0.29–1.09

Notes: OR – odds ratio; p – the significance of differences for control and patients.

Table 3
SNP frequency of FTO, ADRB3, LepR genes in **women (F)**.

Gene	Genotypes/alleles	Genotype/Alleles Frequencies in Groups		χ^2	P	OR*	
	Genotypes	Patients (F) (n = 79)	Control (F) (n = 24)			value	95% CI
FTO (rs9939609)	A/A	0.380	0.375	5.54	0.05	1.02	0.40–2.62
	A/T	0.380	0.583			0.44	0.17–1.11
	T/T	0.241	0.042			7.28	1.05–57.58
	Alleles						
	A	0.570	0.667	1.44	0.23	0.66	0.34–1.30
	T	0.430	0.333			1.51	0.77–2.98
	Genotypes	Patients (n = 28) F	Control (n = 14) F				
ADRB3 (rs4994)	Trp/Trp	0.071	0.143	12.68	0.002	0.46	0.06–3.68
	Trp/Arg	0.000	0.357			0.03	0.00–0.60
	Arg/Arg	0.929	0.500			13.00	2.19–77.04
	Alleles						
	Trp	0.071	0.321	8.92	0.003	0.16	0.04–0.59
	Arg	0.929	0.679			6.16	1.70–22.36
	Genotype	Patients (n = 28) F	Control (n = 14) F				
LepR (rs1137101)	Gln/Gln	0.357	0.214	3.09	0.21	2.04	0.46–9.06
	Gln/Arg	0.464	0.357			1.56	0.42–5.85
	Arg/Arg	0.179	0.429			0.29	0.07–1.22
	Alleles						
	Gln	0.589	0.393	2.89	0.09	2.22	0.88–5.60
	Arg	0.411	0.607			0.45	0.18–1.14

Notes: OR – odds ratio; p – significance of differences for control and patients.
For those women who are at risk of developing obesity, the Trp/Arg and A23525T polymorphisms of the FTO and ADRB3 genes (Table 3) are associated. The homozygous TT genotype FTO (χ^2 = 5.54; p = 0.05) increases the risk of obesity by 7.28 times, the minor 64Arg allele of the ADRB3 gene increases 6.16 times (χ^2 = 8.92; p = 0.003), and Arg64Arg genotype increases by 13.0 times (χ^2 = 12.68; p = 0.002).
Significant differences between patients and controls were not found (Table 4) for rs1137101 of the LepR gene.

Table 4
SNP frequency of FTO, ADRB3, LepR genes in **men (M)**.

Gene	Genotypes/alleles	Genotype/Alleles Frequencies in Groups		χ^2	P	OR*	
	Genotypes	Patients (M) (n = 41)	Control (M) (n = 18)			value	95% CI
FTO (rs9939609)	A/A	0.293	0.167	2.60	0.27	2.07	0.50–8.48
	A/T	0.439	0.667			0.39	0.12–1.25
	T/T	0.268	0.167			1.83	0.44–7.58
	Alleles						
	A	0.512	0.500	0.01	0.9	1.05	0.48–2.30
	T	0.488	0.500			0.95	0.43–2.09
	Genotypes	Patients (n = 12) M	Control (n = 18) M				
ADRB3 (rs4994)	Trp/Trp	0.000	0.278	7.27	0.003	0.10	0.00–1.96
	Trp/Arg	0.000	0.167			0.18	0.01–3.76
	Arg/Arg	1.000	0.556			20.24	1.04–93.65
	Alleles						
	Trp	0.000	0.361	11.06	0.0009	0.04	0.00–0.63
	Arg	1.000	0.639			28.15	1.58–97.35
	Genotype	Patients (n = 12) M	Control (n = 18) M				
LepR (rs1137101)	Gln/Gln	0.250	0.111	1.42	0.49	2.67	0.37–9.06
	Gln/Arg	0.417	0.611			0.45	0.10–2.01
	Arg/Arg	0.333	0.278			1.30	0.27–6.33
	Alleles						
	Gln	0.458	0.417	0.10	0.75	1.18	0.42–3.36
	Arg	0.542	0.583			0.84	0.30–2.39

Notes: OR – odds ratio; p – the significance of differences for control and patients.

loss of visceral fat after the course of the unloading and diet therapy. The analyzed parameters of T23525T homozygotes (Table 5) slightly differ from the reference values (Table 5). For rs4994 ADRB3 gene, similar comparisons were not analyzed, since only two carriers with a normal Trp/Trp genotype and 95% of those surveyed with the Arg/Arg variant were found in the group of people with overweight and obesity.

A correlation analysis between anthropometric features and the genotypes of the studied SNPs had revealed ten significant links only for

Table 5
Analysis of the correlation between the rs1137101 and rs9939609 genotypes and morphophysiological parameters of the examined patients.

Research	Genotype					
	LepR			FTO		
	Gln/Gln M ± δ	Gln/Arg M ± δ	Arg/Arg M ± δ	A/A M ± δ	A/T M ± δ	T/T M ± δ
Weight (kg) to	98.17 ± 5.18	93.01 ± 7.76	102.40 ± 13.04	93.50 ± 3.27	97.89 ± 4.26	113.54 ± 6.56*
P (95%)	*p1-2 < 0,05*	*p2-3 < 0,05*	p1-3 > 0,05	*p4-5 < 0,05*	*p5-6 < 0,005*	*p4-6 < 0,005*
Weight (kg) after	89.14 ± 4.79	84.53 ± 6.98	92.95 ± 12.61	84.93 ± 3.03	89.384 ± 3.96	103.51 ± 6.66**
P (95%)	p1-2 > 0,05	*p2-3 < 0,05*	p1-3 > 0,05	*p4-5 < 0,005*	*p5-6 < 0,005*	*p4-6 < 0,005*
BMI (U) to	34.87 ± 2.04	33.44 ± 2.15	36.04 ± 3.95	32.92 ± 1.09	34.90 ± 1.40	37.85 ± 1.82[a]
P (95%)	p1-2 > 0,05	*p2-3 < 0,05*	p1-3 > 0,05	*p4-5 < 0,005*	*p5-6 < 0,005*	*p4-6 < 0,005*
BMI (unit) after	31.73 ± 1.95	30.42 ± 1.95	32.76 ± 3.85	29.92 ± 1.02	33.95 ± 1.32	35.42 ± 1.72[b]
P (95%)	p1-2 > 0,05	*p2-3 < 0,05*	p1-3 > 0,05	*p4-5 < 0,005*	*p5-6 < 0,005*	*p4-6 < 0,005*
Waist circumference (cm) to	107.69 ± 3.50	103.18 ± 6.25	109.88 ± 9.09	100.83 ± 2.19	105.83 ± 2.92	114.23 ± 4.18[c]
P (95%)	*p1-2 < 0,05*	*p2-3 < 0,05*	p1-3 > 0,05	*p4-5 < 0,005*	*p5-6 < 0,005*	*p4-6 < 0,005*
Waist circumference (cm) after	99.15 ± 3.52	94.75 ± 5.99	101,88 ± 9.27	93.29 ± 2.06	97.00 ± 3.05	106.11 ± 5.22[d]
P (95%)	*p1-2 < 0,05*	*p2-3 < 0,05*	p1-3 > 0,05	*p4-5 < 0,005*	*p5-6 < 0,005*	*p4-6 < 0,005*
Hip circumference (cm) to	117.23 ± 3.94	113.82 ± 4.51	116.25 ± 7.23	113.36 ± 2.83	113.26 ± 2.37	116.81 ± 3.01
P (95%)	*p1-2 < 0,05*	p2-3 > 0,05	p1-3 > 0,05	*p4-5 < 0,005*	*p5-6 < 0,005*	*p4-6 < 0,005*
Thigh circumference (cm) after	111.54 ± 3.95	108.06 ± 3.86	111.38 ± 7.03	108.61 ± 2.15	108.66 ± 2.36	114.42 ± 3.37
P (95%)	*p1-2 < 0,05*	p2-3 > 0,05	p1-3 > 0,05	*p4-5 < 0,05*	*p5-6 < 0,005*	*p4-6 < 0,005*
Visceral fat (kg) to	11.75 ± 1.79	9.25 ± 1.97	15.00 ± 3.21	7.00 ± 2.09	12.40 ± 2.15	13.80 ± 2.03[e]
P (95%)	p1-2 > 0,05	*p2-3 < 0,005*	p1-3 > 0,05	*p4-5 < 0,0001*	*p5-6 < 0,001*	*p4-6 < 0,0001*
Visceral fat (kg) after	10.00 ± 1.70	7.14 ± 1.71	11.60 ± 2.25	6.00 ± 1.84	9.00 ± 1.51	13.00 ± 1.67[f]
P (95%)	*p1-2 < 0,05*	*p2-3 < 0,005*	p1-3 > 0,05	p4-5 < 0,5	*p5-6 < 0,0001*	*p4-6 < 0,0001*
Fat mass (kg) to	41.11 ± 5.74	39.50 ± 5.97	47.62 ± 4.26	39.14 ± 4.15	41.72 ± 3.85	46.91 ± 3.04[g]
P (95%)	p1-2 > 0,05	*p2-3 < 0,05*	p1-3 > 0,05	*p4-5 < 0,05*	*p5-6 < 0,0001*	*p4-6 < 0,0001*
Fat mass (kg) after	37.62 ± 7.52	35.45 ± 7.27	41.52 ± 12.53	34.38 ± 4.37	36.14 ± 4.45	38.27 ± 5.97
P (95%)	p1-2 > 0,05	p2-3 > 0,05	p1-3 > 0,05	*p4-5 < 0,0001*	p5-6 < 0,06	*p4-6 < 0,05*
Muscle mass (kg) to	52.12 ± 5.79	46.42 ± 3.27	57.73 ± 4.81	42.06 ± 2.24	53.76 ± 3.56	54.80 ± 3.079[h]
P (95%)	*p1-2 < 0,001*	*p2-3 < 0,001*	*p1-3 < 0,001*	*p4-5 < 0,005*	*p5-6 < 0,0001*	*p4-6 < 0,0001*
Muscle mass (kg) after	46.64 ± 0.84	42.94 ± 2.76	53.82 ± 4.24	40.86 ± 2.06	49.70 ± 3.16	50.14 ± 3.53[i]
P (95%)	*p1-2 < 0,001*	*p2-3 < 0,001*	*p1-3 < 0,001*	*p4-5 < 0,0001*	p5-6 < 0,5	*p4-6 < 0,05*

Notes: P – significance of differences; 1 - Gln/Gln; 2 - Gln/Arg; 3 - Arg/Arg; 4 – A/A; 5 – A/T; 6 - T/T; *r = +0.410 (p = 0.02); **r = + 0.325 (p = 0.01).
[a] r = +0.222 (p = 0.01).
[b] r = +0.205 (p = 0.007).
[c] r = + 0.426 (p = 0.01).
[d] r = + 0.358 (p = 0.03).
[e] r = +0.541 (p = 0.01).
[f] r = +0.663 (p = 0.002).
[g] r = +0.548 (p = 0.01).
[h] r = +0.587 (p = 0.01).
[i] r = +0.517 (p = 0.01).

the FTO gene, of which six corresponds to weak interconnections ($r < 0.5$; $p \leq 0.05$). The highest correlation coefficient in the group of patients was established between T23525T genotype and indicators of visceral fat, fat and muscle mass before and after the course of unloading and diet therapy (Table 5). As a result, it can be assumed that the mutant homozygous variant rs9939609 is not only associated with an increased risk of obesity but also is a prognostically unfavorable factor for obtaining the final effect from measures aimed for weight correction.

To confirm the hypothesis about the effect of diet and lifestyle on the structure of genes, SNP genes studied before and after the complex health program was typed in patients with obesity. Of the three allelic variants of the genes studied (FTO, ADRB3, LepR), 26 out of 120 (21.6%) patients had changes in the rs9939609 FTO gene (Table 6) after undergoing a course of a low-calorie diet.

It should be noted that processes, associated with the transition of A23525T and T23525T genotypes to the homozygous A23525A variant (Table 6), dominated in 34.6% of cases (from all transitions), where the replacement (transversion) of the purine base T (associated with obesity) to pyrimidine – A (normal or wild allele) occurs.

Gender analysis showed that it was 80% transcription of the pyrimidine and purine bases (A↔T). The phenomenon of transversion to DNA identified in an experimental study indicates the effect of unloading and diet therapy on the replacement of nucleotides in the

Table 6
Comparison of genotype frequencies before and after unloading and diet therapy.

Genotype		Before unloading and diet therapy (n)	After unloading and diet therapy (n)		
			A23525A	A23525T	T23525T
Male (n = 41)	A23525A	12	–	1	–
	A23525T	18	3	–	–
	T23525T	11	–	1	–
Female	A23525A	30	–	2	3
(N = 79)	A23525T	30	4	–	3
	T23525T	19	2	7	–
Total sample	A23525A	42	–	3	3
(n = 120)	A23525T	48	7	–	3
	T23525T	30	2	8	–

structure of genes, in particular, the rs9939609 of the FTO gene. For a more detailed analysis, all transversions were grouped by us into groups based on the general characteristics of the changes presented in Table 7.

The number of transitions in the group with prognostically favorable genotypes is 65.38% of the total number of cases. We analyzed the degree of conjugation of polymorphisms and various anthropometric indicators using the statistical methods Phi and Cramer's V to identify

Table 7
Transverse grouping by the common transition.

Transversion Option	Number of associations
Gt→N; P → N; P→Gt	17
N→ P; N→Gt; Gt → P	9

Notes: N – a normal genotype; Gt – a heterozygote; P – pathological genotype.

possible factors affecting the transversion process. It was found that the presence or absence of obesity (0.755 at p = 0.005) (Table 8) affects the direction of transversion.

According to the presented data and in accordance with the results of the research, obesity in patients is a prognostic unfavorable factor that facilitates the transition to allelic variants associated with the development of pathology (Table 8). The FTO is the normal number of cases or normal BMI. Thus, on the basis of anthropometric patient data, it is possible to predict the probability of the transition of the rs9939609 genotype after a course of a low-calorie diet.

4. Discussion

The search and improvement of the methodological base for the early diagnosis and treatment of obesity is one of the urgent tasks of modern clinical medicine. Therefore, research, related to the study of molecular genetic mechanisms of regulation of energy processes in the body and the identification of informative markers, can significantly improve this process.

One of the promising directions in the typing of genes, involved in metabolic processes, is the adrenoreceptor – β3 from the family of receptors, associated with G-proteins, and expressed in adipose tissue, smooth muscle of blood vessels, gastrointestinal tract and gall bladder (Anthony et al., 1998; Shihara et al., 1999). The main physiological effect is due to the participation in the control of lipolysis and thermogenesis. Activation of the ADRB3 receptors through Gs proteins triggers adenylate cyclase and formation of secondary messenger cAMP, which stimulates lipolysis in white adipose tissue and heat production in brown adipose tissue (Bachman et al., 2002). To determine the role of ADRB3 in the regulation of lipolysis, in response to a change in diet (Bachman and others 2002), a line of mice with the knockout gene ADRB3 (Walston et al., 1995) was developed, in which, unlike the wild type, obesity develops in response to the hypercaloric diet with high content of lipids. These studies confirm the key role of ADRB3 in maintaining energy balance by changing the intensity of heat transfer from the body and protecting against dyslipidemia.

When sequencing the ADRB3 gene, a single missense mutation with the replacement of tryptophan (Trp) to arginine (Arg) in position 64 (rs4994) of the first coding exon was detected at the junction of the transmembrane domain and the intracellular loop of the receptor (Candelore et al., 2007; Piétri-Rouxel et al., 1997; Kurokawa et al., 2008). Fifty-eight papers, related to obesity, were submitted in the network "HuGE Literature Finder" on rs4994 SNP. Four large meta-analyses (The National Center for Biotechnology Information, 2019) did not confirm the association of rs4994 with the risk of obesity; however, a significant relationship was established between 64Arg and metabolic disturbances, mainly in Asian populations of China, Japan, and Korea (Kurokawa et al., 2008; Oizumi et al., 2001; Mo et al., 2007; Yamakita et al., 2010; Zhu et al., 2010; Yoshihara et al., 2014; Liu et al., 2015).

In contrast to the cited works, the association of the Tpr64Arg gene polymorphism with obesity, both for the homozygous mutant Arg/Arg genotype (p = 0.0001) and the 64Arg allele (p < 0.0001), was established with a high degree of confidence (p « 0.001) in our study. The frequency of 64Arg alleles in the group of patients is 95%; it significantly differs (from 2% to 23%) from the results of other studies (The National Center for Biotechnology Information, 2019). The presence of mutant allele increases the risk of obesity by six times, and homozygous Arg64Arg increases by 13 times. The dependence of the main anthropometric parameters on the identified genotypes was not analyzed due to the apparent dominance of the mutant Arg64Arg of the homozygous variant of 95.0%. The frequency of the Arg64Arg genotype in the control group (53.1%) of the inhabitants of the North Caucasus differs from the average statistical data in the world according to the results of the “1000 Genomes Project” (4.7%–21.6%) (Krief et al., 1993). The high frequencies of 64Arg alleles and Arg64Arg of the ADRB3 genotype, obtained in our study, may be due to the specifics of the sample of subjects or the ethnogenetic characteristics of the population living in the region.

The reliable association of the 64Arg allele of ADRB3 with an increased risk of developing hyperglycemia to a greater extent than with obesity (Shcherbakova et al., 2010; Kochetova et al., 2015, 2017; Baturin et al., 2012a, 2014, 2016a; Borodin and Gapparova, 2016) was confirmed by studies in Russia. The frequency of 64Arg polymorphism in some regions varies in a narrow range of values of 9.6%–22.2% and is comparable with foreign data. However, it should be noted that in 2012, A.K. Baturin, A.V. Pogozheva, E.I. Sorokina, and others did not reveal statistically significant differences between control and patients; and 64Arg association with obesity was confirmed by the same authors in 2016 (A.K. Baturin, A.V. Pogozheva, E.I. Sorokina, et all. 2016). This fact may be due to both the number of participants in the experiment and the ethnogenetic features of the persons examined. According to S.V. Borodin et al. (2016), rs4994 of the ADRB3 gene is not involved in the pathological process. In the work of T.V. Kochetova, and others (2015), on the contrary, a relationship of the prognostically unfavorable 64Arg ADRB3 polymorphism in women of the Tatar population (Kochetova et al., 2015) was shown. However, we found that rs4994 has a more significant prognostic significance for men: the carriage of

Table 8
Conjugation analysis between FTO genotypes and obesity.

Contingency table							
Combination of genotypes		Obesity					Total
		No obesity	1° obesity	2° obesity	3° obesity	Extraweight	
Gt→N; P → N; P→Gt (n = 17)	Frequency	10	2	2	0	3	17
	Expected frequency	6.5	3.3	3.9	1.3	2.0	17.0
N→ P; N→Gt;	Frequency	0	3	4	2	0	9
Gt → P (n = 9)	Expected frequency	3.5	1.7	2.1	0.7	1.0	9.0
Total	Frequency	10	5	6	2	3	26
	Expected frequency	10.0	5.0	6.0	2.0	3.0	26.0
Rated at rated	Phi	0.755					
		p = 0.005					
	Cramer's V	0.755					
		p = 0.005					

the homozygous Arg64Arg genotype increases the risk of developing obesity by a factor of 20 times, whereas in women it increases only 13 times. Based on the presented materials, we can make a preliminary conclusion about the effectiveness of using this polymorphism as an early marker of metabolic disorders, especially for men of the south of Russia.

Another analyzed polymorphism, associated with an increased risk of obesity, is the missense mutation of the LEPR gene, due to a change in the adenine nucleotide to guanine at 668 position (rs1137101) in 4 exon, followed by the replacement of the glutamine amino acid with arginine at 223 positions (Gln223Arg/Q223R) of the protein product, which affects the increase in food intake and the development of obesity (The National Center for Biotechnology Information, 2019; The Allele Frequency Database, 2019; Zhang et al., 1994; Inui, 1999; Chung et al., 1997; Chagnon et al., 1999; Chagnon et al., 2000).

The results of 124 independent studies on the association of the L2R gene polymorphism with diseases, including 46 with the risk of developing obesity, were published in international information databases. However, most authors deny the association of rs1137101 with overweight (Paracchini et al., 2000; Gallicchio et al., 2009; Pyrzak et al., 2009; Constantin et al., 2010; Angel-Chávez et al., 2012; Dias et al., 2012; Rustemoglu et al., 2012; Görmüş et al., 2014; Al-Azzam et al., 2014; Ng et al., 2014; Gajewska et al., 2016; An et al., 2016), which is confirmed by the results of two large meta-analyses conducted in 2002 and 2011 involving 15,432 people (case/control) and did not reveal a significant relationship between Q223R SNP and metabolic disturbance (Heo et al., 2002; Bender et al., 2011). However, in separate studies, carried out in Turkey, Malaysia, Romania, Brazil, Tunisia, England and the Netherlands, it was shown that 223R polymorphism is associated with an increased risk of obesity, since the carriage of the minor allele correlates with elevated cholesterol, triglycerides and significantly reduces insulin sensitivity (Quinton et al., 2001; Duarte et al., 2007; van der Vleuten et al., 2006; Boumaiza et al., 2012; Becer et al., 2013; Kasim et al., 2016; Wauters et al., 2002; Sook-Ha and Yee-How, 2014; Hassan et al., 2017; Berezina et al., 2015; Nikolaev et al., 2014; Zyablitsev et al., 2016; Morozova et al., 2014).

The association of rs1137101 polymorphism with the risk of obesity in residents of southern Russia is also not confirmed in our studies, but in other domestic studies (Takahashi-Yasuno et al., 2003; Furusawa et al., 2010) a significant association of 223R with lipid metabolism, correlated with cholesterol, triglycerides, and other biochemical blood parameters were found. Therefore, we analyzed the relationship of genotypes with weight, BMI, waist circumference and other main anthropometric indicators. Carrying a homozygous mutant variant of the Arg/Arg variant, compared to the heterozygous Gln/Arg genotype, is associated with a statistically significant impairment of physiological parameters.

When analyzing the results of studies in comparison with the data of other authors (Paracchini et al., 2000; Gallicchio et al., 2009; Pyrzak et al., 2009; Constantin et al., 2010; Angel-Chávez et al., 2012; Dias et al., 2012; Rustemoglu et al., 2012; Görmüş et al., 2014; Al-Azzam et al., 2014; Ng et al., 2014; Gajewska et al., 2016; An et al., 2016), it should be noted that the carriers of Gln223Arg variant of the LEPR gene before and after RTT revealed the lowest anthropometric indicators, which determines the effectiveness of treatment. However, the significance of rs1137101, as a genetic marker in the diagnosis of obesity, remains open.

Search for markers associated with the development of metabolic processes in the body, started with the gene "Fat mass and obesity-associated protein" (FTO), which is expressed in almost all tissues of the body (brain, muscles, adipose tissue, adrenal glands, pancreas) and is one from promising objects in the study of the genetics of obesity (Scuteri et al., 2007b; Gulati et al., 2013; Wu et al., 2010; Hubacek et al., 2012; Liu et al., 2010).

According to “The Allele Frequency Database” (date of appeal 2019), 138 mutations were typed in the FTO gene, most of which have no functional significance. In clinical medicine, rs9939609 polymorphism with nucleotide substitution (SNP) of adenine (A) for thymine (T) in position 23525 of the first intron is the most promising. The results of 438 scientific studies on the association of the A23525T polymorphism with various diseases, of which 234 are associated with obesity (Genome-wide association studies, 2019; The National Center for Biotechnology Information, 2019), were published in the database “HuGE Literature Finder” (2010–2019). Multicenter studies have confirmed the role of the homozygous AA genotype and the A allele of the FTO gene in the development of obesity regardless of the ethnicity of the individuals studied (Quan et al., 2015; Mačeková et al., 2015; Tanofsky-Kraff et al., 2009; Lingwei et al., 2016; Lurie et al., 2011a, 2011b; Deliard et al., 2013; Smemo et al., 2014; Stratigopoulos et al., 2014; Yang et al., 2014; Kilpeläinen et al., 2011; Livingstone et al., 2016; Korelskaya et al., 2014). The prognostically unfavorable allele is predominantly associated with central or abdominal obesity.

In domestic studies (Zavyalova et al., 2011; Nasibulina et al., 2012; Baturin et al., 2011, 2012b, 2016b, 2017), the association of the FTO gene with an increased body mass index was also noted, but in contrast to the cited works, the homozygous T23525T genotype is associated with obesity ($\chi^2 = 7.20$ $p = 0.03$) in the residents of the Republic of Adygeya, which increases the risk of obesity by 3 times, which is much higher than the average indicators (OR = 1.2–1.8) in the world and the Russian Federation (Mačeková et al., 2015; Tanofsky-Kraff et al., 2009; Lingwei et al., 2016; Lurie et al., 2011a, 2011b; Deliard et al., 2013; Smemo et al., 2014; Stratigopoulos et al., 2014; Yang et al., 2014; Kilpeläinen et al., 2011; Livingstone et al., 2016; Korelskaya et al., 2014; Zavyalova et al., 2011; Nasibulina et al., 2012; Baturin et al., 2011, 2012b, 2016b, 2017). It should be noted that the identified gender differences in the distribution of Rs9939609 and the connection with obesity mainly in women ($\chi^2 = 5.54$; $p = 0.05$) are not reflected in the analyzed works from the database “HuGE Literature Finder” (The National Center for Biotechnology Information, 2019).

An important aspect of experimental work is the establishment of a relationship between phenotypic traits and genotypes of the examined individuals (Arkadianos et al., 2007; Celis-Morales et al., 2016; Petkeviciene et al., 2016; Camp and Trujillo, 2014). Taking into account the genetic features in the present work, the correlation between the T23525T genotype and the indicators of visceral fat, fat and muscle mass of the body has been proved until and after the course of the complex health program. As a result, it can be assumed that the mutant homozygous variant rs9939609 is not only associated with an increased risk of obesity but also is a prognostically unfavorable factor for carrying out measures aimed for weight correction. However, based on literature data (Celis-Morales et al., 2016; Petkeviciene et al., 2016; Camp and Trujillo, 2014), as well as own results, it was found that the presence of mutation does not interfere with the process of normalizing the physiological parameters of the body as a result of therapeutic effects.

Many of the effects of the hypocaloric diet, according to some authors (Grayson et al., 2014; Toperoff et al., 2015; Pitman and Borgland, 2015), are carried out by epigenetic regulation of the expression of genes responsible for DNA repair. There is a study proving the effect of whole cow milk on the epigenetic processes in the FTO gene through exosomal miR-29 (Melnik, 2015).

Therefore, we have typed SNPs of the FTO, ADRB3, LepR genes in the genomic DNA of patients before and after the complex health program. Despite the fact that three genes were examined in the experiment, the transversion phenomenon was found only for rs9939609 FTO. The results confirmed by a blind method in an independent laboratory with the observance of similar conditions suggest that in the process of carrying out complex treatment in 26 (21.6%) of 120 examined patients there had been a change in the rs9939609 genotype. In 34.6% of all transversion cases, a reversal of mutations was observed, due to the T→A nucleotide substitution and the transfer of the A23525T, T23525T genotypes to the homozygous A23525A variant, i.e.

the return of the gene to its original state. We have also established a high degree of correlation between the rs9939609 polymorphisms and obesity, so we assume that the presence of pathology is a prognostically unfavorable factor determining the direction of transversion. It should be noted that patients with an unfavorable genotype transition, gained about 5.16 ± 1.8 kg after 6–18 months and patients with a favorable transverse version retained their weight (−0.3 ± 1.8 kg; $p < 0.05$) after the complex health program.

It was revealed that age might be another possible factor affecting the transversion process. Based on the obtained data, a tendency was revealed which is related to the fact that the direction of transfer was due to the transition to unfavorable genetic variants in all the examined persons at the age of (56.5 ± 1.7). Thus, perhaps, age is an additional factor that can affect this phenomenon under unloading and diet therapy.

One of the possible mechanisms of the established phenomenon is the fact that specific reactions contributing to the destabilization of the FTO gene, activation of the mechanisms promoting the transversion process, caused by changes in the functional activity of nuclear regulatory systems: NF-kB and p53, develop in some patients against the background of the complex health program. Processes at the eustress level can enhance reparative mechanisms with the restoration of mutated genes. With distress, modifications of nucleotides are possible, i.e. mutation of individual DNA genes. Probably, this can explain the multidirectionality of the transversion of point mutations in the FTO gene under conditions of caloric restriction in our research. Thus, the results of our research and model experiments in vivo V.D. Longo and others (2010–2016) (Longo and Fontana, 2010; Longo et al., 2013, 2014, 2015) confirm the possible effect of a low-calorie diet, exercise, physiotherapy procedures on the structure of genes. However, further research is needed to clarify this mechanism.

5. Conclusion

The results of this study are not only theoretical but also of practical importance since they complement and expand the fundamental knowledge about the role of gene polymorphisms in the development of obesity, the effect of partial food deprivation and health measures on the nucleotide sequence of genes. However, data on transversion and theoretical assumptions, expressed in the paper on the effect of the complex health program on the rs9939609 FTO gene, do not exclude a multi-level regulation of the processes of reparation and epigenetic activation of genes. Therefore, population studies of the phenomenon of A23525T transversion of the polymorphism of the FTO gene can broaden the understanding of the molecular-genetic mechanisms for the development and correction of body weight in obesity.

References

Al-Azzam, S.I., Khabour, O.F., Alzoubi, K.H., 2014. The effect of leptin promoter and leptin receptor gene polymorphisms on lipid profile among the diabetic population: modulations by atorvastatin treatment and environmental factors. J. Endocrinol. Investig. 37 (9), 835–842. https://doi.org/10.1007/s40618-014-0113-6.

An, B.Q., Lu, L.L., Yuan, C., et al., 2016. Leptin receptor gene polymorphisms and the risk of non-alcoholic fatty liver disease and coronary atherosclerosis in the Chinese han population. Hepat. Mon. 16 (4), 1–7. https://doi.org/10.5812/hepatmon.35055.

Angel-Chávez, L.I., Tene-Pérez, C.E., Castro, E., 2012. Leptin receptor gene K656N polymorphism is associated with low body fat levels and elevated high-density cholesterol levels in Mexican children and adolescents. Endocr. Res. 37, 124–134. https://doi.org/10.3109/07435800.2011.648360.

Anthony, A., Schepelmann, S., Guillaume, J.L., et al., 1998. Localization of the beta(β)3-adrenoceptor in the human gastrointestinal tract: an immunohistochemical study. Aliment. Pharmacol. Ther. 12, 519–525. https://doi.org/10.1016/j.ejphar.2015.02.031.

Arkadianos, I., Valdes, A.M., Efstathios, M., et al., 2007. Improved weight management using genetic information to personalize a calorie controlled diet. J. Nutr. 6, 229–237. https://doi.org/10.1186/1475-2891-6-29.

Bachman, E.S., Dhillon, H., Zhang, C.-Y., et al., 2002. Beta-AR signaling required for diet-induced thermogenesis and obesity resistance. Science 297, 843–845. https://doi.org/10.1126/science.1073160.

Baturin, A.K., Pogozheva, A.V., Sorokina, E.Yu, et al., 2011. Study of the polymorphism rs 9939609 of the FTO gene in persons with overweight and obesity. J. Nut. issues 3, 13–17 (in Russian).

Baturin, A.K., Pogozheva, A.V., Sorokina, E.I., 2012a. The Trp64Arg polymorphism of beta3-adrenoreceptor gene study in persons with overweight and obesity. Vopr. Pitan. 81 (2), 23–27 (in Russian).

Baturin, A.K., Pogozheva, A.V., Sorokina, E.Yu, et al., 2012b. Study of the polymorphism of the rs9939609-FTO gene in the population of the Sverdlovsk Region. J. Nut. Issues: Sci. Prac. 5, 28–32 (in Russian).

Baturin, A.K., Sorokina, E.Yu, Pogozheva, A.V., 2014. Regional features of polymorphism of genes associated with obesity (rs9939609 of the FTO gene and Trp64Arg of the ADRB3 gene) in the Russian population. Nutr. Issues: Sci. Practical. Mag. 2, 35–41 (in Russian).

Baturin, A.K., Sorokina, E.Yu, Pogozheva, A.V., 2016a. Study of the combined effect of genetic polymorphisms of the rs9939609 gene FTO and rs4994 of the gene ADRB3 on the risk of developing obesity. Nutr. Issues: Sci. Practical. Mag. 4, 29–34 (in Russian).

Baturin, A.K., Pogozheva, A.V., Sorokina, E.Yu, et al., 2016b. A study of the combined effect of the genetic polymorphisms rs9939609 of the FTO gene and rs4994 of the ADRB3 gene on the risk of obesity development. J. Nut. issues: Sci. Prac. 4, 29–34 (in Russian).

Baturin, A.K., Pogozheva, A.V., Sorokina, E.Yu, et al., 2017. Study of the association of polymorphisms rs993609 of the FTO gene and rs659366 of the UCP2 gene with obesity in the population of the Arctic zone of the Russian Federation. J. Nut. issues: Sci. Prac. 3, 32–39 (in Russian).

Becer, E., Mehmetçik, G., Bareke, H., 2013. Association of leptin receptor gene Q223R polymorphism on lipid profiles in comparison study between obese and non-obese subjects. Gene 529 (1), 16–20. https://doi.org/10.1016/j.gene.2013.08.003.

Bender, N., Allemann, N., et al., 2011. Association between variants of the leptin receptor gene (LEPR) and overweight: a systematic review and an analysis of the CoLaus study. PLoS One 6 (10), 25–36. https://doi.org/10.1371/journal.pone.0026157.

Berezina, A., Belyaeva, O., Berkovich, et al., 2015. Risk factors, and genetic traits in metabolically healthy and unhealthy obese individuals. BioMed Res. Int. 45, 54–58 (in Russian).

Borodin, C.B., Gapparova, K.M., 2016. Genetic predictors of obesity. Obes. Metab. 13 (2), 7–13 (in Russian).

Boumaiza, I., Omezzine, A., Rejeb, J., et al., 2012. Relationship between leptin G2548A and leptin receptor Q223R gene polymorphisms and obesity and metabolic syndrome risk in Tunisian volunteers. Genet. Test. Mol. Biomark. 16, 726–733. https://doi.org/10.1089/gtmb.2011.0324.

Camp, K.M., Trujillo, E., 2014. Position of the academy of nutrition and dietetics: nutritional genomics. J. Acad. Nutr. Diet. 114 (2), 299–312. https://doi.org/10.1016/j.jand.2013.12.001.

Candelore, M.R., Deng, L., Tota, L.M., et al., 2007. Pharmacological characterization of a recently described human beta 3-adrenergic receptor mutant. Endocrinology 137 (6), 2638–2641. https://doi.org/10.1210/endo.137.6.8641219.

Celis-Morales, C., Marsaux, C.F., Livingstone, K.M., et al., 2016. Physical activity attenuates the effect of the FTO genotype on obesity traits in European adults: the Food4Me study. Obesity 24 (4), 962–979. https://doi.org/10.1002/oby.21422.

Chagnon, Y.C., Chung, W.K., Pérusse, L., et al., 1999. Linkages and associations between the leptin receptor (LEPR) gene and human body composition in the Québec Family Study. Int. J. Obes. Relat. Metab. Disord. 23 (3), 278–286. https://doi.org/10.1038/sj.ijo.0800809.

Chagnon, Y.C., Wilmore, J.H., Borecki, I.B., et al., 2000. Associations between the leptin receptor gene and adiposity in middle-aged Caucasian males from the HERITAGE family study. J. Clin. Endocrinol. Metab. 85 (1), 29–34. https://doi.org/10.1210/jcem.85.1.6263.

Chung, W.K., Power-Kehoe, L., Chua, M., et al., 1997. Exonic and intronic sequence variation in the human leptin receptor gene (LEPR). Diabetes 46 (9), 1509–1511. https://doi.org/10.2337/diab.46.9.1509.

Constantin, A., Costache, G., Sima, A.V., 2010. Leptin G-2548A and leptin receptor Q223R gene polymorphisms are not associated with obesity in Romanian subjects. Biochem. Biophys. Res. Commun. 391 (1), 282–296. https://doi.org/10.1016/j.bbrc.2009.11.050.

Deliard, S., Panossian, S., Mentch, F.D., et al., 2013. The missense variation landscape of FTO, MC4R, and TMEM18 in obese children of African Ancestry. Obesity 21 (1), 159–163. https://doi.org/10.1002/oby.20147.

Dias, N.F., Fernandes, A.E., Melo, M.E., et al., 2012. Lack of mutations in the leptin receptor gene in severely obese children. Arq. Bras. Endocrinol. Metabol. 56 (3), 178–183. https://doi.org/10.1590/S0004-27302012000300005.

Duarte, S.F., Francischetti, E.A., Genelhu, V.A., 2007. LEPR p.Q223R, beta3-AR p.W64R and LEP c.-2548G > A gene variants in obese Brazilian subjects. Genet. Mol. Res. 6, 1035–1043.

Furusawa, T., Naka, I., Yamauchi, T., et al., 2010. The Q223R polymorphism in LEPR is associated with obesity in Pacific Islanders. Hum. Genet. 127 (3), 287–294. https://doi.org/10.1007/s00439-009-0768-9.

Gajewska, J., Kuryłowicz, A., Mierzejewska, E., et al., 2016. Complementary effects of genetic variations in LEPR on body composition and soluble leptin receptor concentration after 3-month lifestyle intervention in prepubertal obese children. Nutrients 8 (6), 125–132. https://doi.org/10.3390/nu8060328.

Gallicchio, L., Chang, H.H., Christo, D.K., et al., 2009. Single nucleotide polymorphisms in obesity-related genes and all-cause and cause-specific mortality: a prospective cohort study. BMC Med. Genet. 10, 203–225. https://doi.org/10.1186/1471-2350-10-103.

Genome-wide association studies https://www.ebi.ac.uk/gwas/, Accessed date: 23 January 2019.

Görmüş, U., Tımırcı Kahraman, Ö., Toptaş, B., et al., 2014. Leptin and leptin receptor polymorphisms are related to body mass index in a Turkish population. Turk. J. Med.

Sci. 44 (5), 809–813. https://doi.org/10.3906/sag-1211-80.
Grayson, S.E., Ponce De Leon, F.A., Muscoplat, C.C., et al., 2014. Epigenetics: understanding how our choices lead to our diseases. J. Clin. Case Rep. 4, 447. https://doi.org/10.4172/2165-7920.1000447.
Gulati, P., Cheung, M.K., Antrobus, R., et al., 2013. Role for the obesity-related FTO gene in the cellular sensing of amino acids. J. Proc. Nat. Acad. Sci. U. S. A. 110, 2557–2562. https://doi.org/10.1073/pnas.1222796110.
Hassan, N.E., El-Masry, S.A., Zarouk, W., et al., 2017. Obesity phenotype in relation to gene polymorphism among samples of Egyptian children and their mothers. Genes Dis. 5 (2), 150–157. https://doi.org/10.1016/j.gendis.2017.12.004.
Heo, M., Leibel, R.L., Fontaine, K.R., et al., 2002. A meta-analytic investigation of linkage and association of common leptin receptor (LEPR) polymorphisms with body mass index and waist circumference. Int. J. Obes. Relat. Metab. Disord. 26 (5), 640–646. https://doi.org/10.1038/sj.ijo.0801990.
Hindorff, L.A., et al., 2009. Potential etiologic and functional implications of genome-wide association loci for human diseases and traits. Proc. Natl. Acad. Sci. U. S. A 106, 9362–9367. https://doi.org/10.1073/pnas.0903103106.
Hubacek, J., Ceska, R., Dlouha, D., 2012. Effect of FTO and MC4R variants on BMI decrease in children. Obes. Facts 5, 165–172. https://doi.org/10.1016/j.clinbiochem.2012.11.017.
HuGE Literature Finder https://phgkb.cdc.gov/PHGKB/startPagePubLit.action, Accessed date: 15 April 2019.
Inui, A., 1999. Feeding and body-weight regulation by hypothalamic neuropeptides—mediation of the actions of leptin. Trends Neurosci. 22 (2), 62–67. https://doi.org/10.1016/S0166-2236(98)01292-2.
Kasim, N.B., Huri, H.Z., Vethakkan, S.R., 2016. Genetic polymorphisms associated with overweight and obesity in uncontrolled Type 2 diabetes mellitus. Biomark. Med. 10 (4), 403–4715. https://doi.org/10.2217/bmm-2015-0037.
Kilpeläinen, T.O., Qi, L., Brage, S., et al., 2011. Physical activity attenuates the influence of FTO variants on obesity risk: a meta-analysis of 218,166 adults and 19,268 children. PLoS One 11, 235–244. https://doi.org/10.1371/journal.pmed.1001116.
Kochetova, O.V., Viktorova, T.V., Mustafina, O.E., 2015. Genetic association of ADRA2A and ADRB3 genes with metabolic syndrome among the tatars. Genetika 51 (7), 830–834 (in Russian).
Kochetova, O.V., Korytina, G.F., Akhmadishin, L.Z., Viktorova, T.V., 2017. Association of polymorphic variants of adrenergic receptor genes (ADRA2A, ADRB3) with obesity in women of Tatar ethnicity. 3, 21–26 (in Russian).
Korelskaya, N.A., Berezina, A.V., Bazhenova, E.A., 2014. The gene associated with fat mass and obesity, and its role in the formation of metabolic syndrome components. J. Bul. Rus. Acad. Nat. Sci. 18 (2), 109–118. (in Russian). https://doi.org/10.1016/j.tig.2010.02.006.
Krief, S., Lönnqvist, F., Raimbault, S., et al., 1993. Tissue distribution of beta 3-adrenergic receptor mRNA in man. J. Clin. Investig. 91 (1), 344–349. https://doi.org/10.1172/JCI116191.
Kurokawa, N., Young, E.H., Oka, Y., et al., 2008. The ADRB3 Trp64Arg variant and BMI: a meta-analysis of 44 833 individuals. Int. J. Obes. (Lond) 32 (8), 1240–1249. https://doi.org/10.1038/ijo.2008.90.
Kuzin, S., Karmanov, M., 2016. Obesity as a specific obstacle sustainable social and economic development. J. Econ. Econ. Sci. 7, 277–282 (in Russian).
Lingwei, X., Hongyu, W., An, P., et al., 2016. FTO genotype and weight loss in diet and lifestyle interventions: a systematic review and meta-analysis. Am. J. Clin. Nutr. 103, 1–9. https://doi.org/10.3945/ajcn.115.123448.
Liu, G., Zhu, H., Lagou, V., et al., 2010. FTO variant rs9939609 is associated with body mass index and waist circumference, but not with energy intake or physical activity in European- and African-American youth. BMC Med. Genet. 11, 1587–1595. https://doi.org/10.1186/1471-2350-11-57.
Liu, J., Zhang, B., Li, M., et al., 2015. Study on relationship between Trp64Arg polymorphism of β3-adrenergic receptor gene and obesity and blood lipids. Zhonghua Yi Xue Za Zhi 95 (20), 1558–1562. https://doi.org/10.3760/cma.j.issn.0376-2491.2015.20.003.
Livingstone, K.M., Celis-Morales, C., Marsaux, C.F., et al., 2016. Profile of European adults interested in internet-based personalised nutrition: the Food4Me study. Eur. J. Nutr. 55 (2), 759–769. https://doi.org/10.1007/s00394-015-0897-y.
Longo, V.D., Fontana, L., 2010. Calorie restriction and cancer prevention: metabolic and molecular mechanisms. Trends Pharmacol. Sci. 31 (2), 89–98. https://doi.org/10.1016/j.tips.2009.11.004.
Longo, V.D., Fontana, L., Adelaiye, R.M., et al., 2013. Dietary protein restriction inhibits tumor growth in human xenograft models. Oncotarget 4 (12), 2451–2461. https://doi.org/10.18632/oncotarget.1586.
Longo, V.D., Fontana, L., Kennedy, B.K., et al., 2014. Medical research: treat ageing. Nature 511 (7510), 405–417. https://doi.org/10.1038/511405a.
Longo, V.D., Antebi, A., Bartke, A., et al., 2015. Interventions to slow aging in humans: are we ready? Aging Cell 14 (4), 497–510. https://doi.org/10.1111/acel.12338.
Lurie, G., Gaude, M.M., Spurdle, A.B., et al., 2011a. The obesity-associated polymorphisms FTO rs9939609 and MC4R rs17782313 and endometrial cancer risk in non-hispanic white women. J. PLoS One 6 (2), 85–97. https://doi.org/10.1371/journal.pone.0016756.
Lurie, G., Gaude, M.M., Spurdle, A.B., et al., 2011b. The obesity-associated polymorphisms FTO rs9939609 and MC4R rs17782313 and endometrial cancer risk in non-hispanic white women. J. PLoS One 6 (2), 85–97. https://doi.org/10.1371/journal.pone.0016756.
Mačeková, S., Bernasovský, I., Gabriková, D., et al., 2015. Association of the FTO rs9939609 polymorphism with obesity in Roma/Gypsy population. Am. J. Phys. Anthropol. 147 (1), 30–34. https://doi.org/10.1002/ajpa.21604.
Melnik, B.C., 2015. Linking diet to acne metabolomics, inflammation, and comedogenesis: an update. Clin. Cosmet Investig. Dermatol. 8, 371–388. https://doi.org/10.2147/CCID.S69135.
Mo, W., Zhang, G.G., Yang, T.L., et al., 2007. The genetic polymorphisms of beta3-adrenergic receptor (AR) Trp64Arg and beta2-AR Gln27Glu are associated with obesity in Chinese male hypertensive patients. Clin. Chem. Lab. Med. 45 (4), 493–498. – 2007. https://doi.org/10.1515/CCLM.2007.089.
Morozova, A.V., Maltseva, N.V., Gorbatovsky, YaA., et al., 2014. An association study of the Gln223Arg polymorphism of the leptin receptor gene with obesity and non-alcoholic fatty liver disease. RZGGK 3, 49–57 (in Russian).
Nasibulina, M.S., Shagimardanova, R.R., Borisova, A.V., 2012. Association of polymorphism of the FTO gene with overweight in the Russian. J. Kazan Med. 5, 823–826 (in Russian).
Ng, Z.Y., Veerapen, M.K., Hon, W.M., 2014. Association of leptin/receptor and TNF-α gene variants with adolescent obesity in Malaysia. Pediatr. Int. 56 (5), 689–697. https://doi.org/10.1111/ped.12336.
Nikolaev, I.V., Mulyukova, R.V., Kayumova, L.R., 2014. Analysis of the interaction of alleles of lipid exchange genes in dyslipidemia. Vavilovsky J. Genet. Breed. 4, 856–866 (in Russian).
Oizumi, T., Daimon, M., Saitoh, T., et al., 2001. Genotype Arg/Arg, but not Trp/Arg, of the Trp64Arg polymorphism of the beta(3)-adrenergic receptor is associated with type 2 diabetes and obesity in a large Japanese sample. Diabetes Care 24 (9), 1579–1583. https://doi.org/10.2337/diacare.24.9.1579.
Paracchini, V., Pedotti, P., Taioli, E., et al., 2000. Genetics of leptin and obesity: a HuGE review. Am. J. Epidemiol. 162, 101–114. https://doi.org/10.1093/aje/kwi174.
Petkeviciene, J., Smalinskiene, A., Klumbiene, J., et al., 2016. Physical activity, but not dietary intake, attenuates the effect of the FTO rs9939609 polymorphism on obesity and metabolic syndrome in Lithuanian adult population. Public Health 135, 23–29. https://doi.org/10.1016/j.puhe.2016.02.009.
Piétri-Rouxel, F., John Manning, B., Gros, J., et al., 1997. The biochemical effect of the naturally occurring Trp64–>Arg mutation on human beta3-adrenoceptor activity. Eur. J. Biochem. 247 (3), 1174–1179. https://doi.org/10.1111/j.1432-1033.1997.01174.x.
Pitman, K.A., Borgland, S.L., 2015. Changes in mu-opioid receptor expression and function in the mesolimbic system after long-term access to a palatable diet. Pharmacol. Ther. 154, 110–119. https://doi.org/10.1016/j.pharmthera.2015.07.005.
Puzyrev, V.P., Freidin, M.B., Kucher, A.N., 2007. Genetic diversity of population and human diseases Tomsk. Print Manufactory 320 (in Russian).
Pyrzak, B., Wisniewska, A., Kucharska, A., et al., 2009. No association of LEPR Gln223Arg polymorphism with leptin, obesity or metabolic disturbances in children. Eur. J. Med. Res. 14, 201–204. https://doi.org/10.1186/2047-783X-14-S4-201.
Quan, L.L., Wang, H., Tian, Y., et al., 2015. Association of fat-mass and obesity-associated gene FTO rs9939609 polymorphism with the risk of obesity among children and adolescents: a meta-analysis. Eur. Rev. Med. Pharmacol. Sci. 19 (4), 614–623.
Quinton, N.D., Lee, A.J., Ross, R.J., et al., 2001. A single nucleotide polymorphism (SNP) in the leptin receptor is associated with BMI, fat mass and leptin levels in postmenopausal Caucasian women. Hum. Genet. 108 (3), 233–236. https://doi.org/10.1007/s004390100468.
Rustemoglu, A., Sahin, S., Tasliyurt, T., 2012. Relationship between obesity and Leptin (G-2548A) and Leptin receptor (668A > G (Q223R)) gene polymorphisms in Turkish population. Endocr. Abstr. 45, 1273.
Scuteri, A., Sanna, S., Chen, W.M., et al., 2007b. Genome-wide association scan shows genetic variants in the FTO gene are associated with obesity-related traits. J. PLoS Genet. 3 (7), 115–128. https://doi.org/10.1371/journal.pgen.0030115.
Shcherbakova, Miu, Sinitsin, P.A., Poriadina, G.I., et al., 2010. Correlation of metabolic syndrome clinical signs and genetic determinants at children with obese Eksp Klin Gastroenterol. 7, 6–11 (in Russian).
Shihara, N., Yasuda, K., Moritani, T., et al., 1999. The association between Trp64Arg polymorphism of the beta-3-adrenergic receptor and autonomic nervous system activity. J. Clin. Endocr. Metab. 84, 1623–1627. https://doi.org/10.1210/jcem.84.5.5701.
Smemo, S., Tena, J.J., Kim, K.H., et al., 2014. Obesity-associated variants within FTO form long-range functional connections with IRX3. J. Nat. 507, 371–385. https://doi.org/10.1038/nature13138.
Sook-Ha, F., Yee-How, S., 2014. Leptin and leptin receptor gene polymorphisms and their association with plasma leptin levels and obesity in a multi-ethnic Malaysian suburban population. J. Physiol. Anthropol. 33 (15). https://doi.org/10.1186/1880-6805-33-15.
Stratigopoulos, G., Martin Carli, J.F., O'Day, D.R., et al., 2014. Hypomorphism for RPGRIP1L, a ciliary gene vicinal to the FTO locus, causes increased adiposity in mice. J. Cell Metab. 19 (5), 767–779. https://doi.org/10.1016/j.cmet.2014.04.009.
Takahashi-Yasuno, A., Masuzaki, H., Miyawaki, T., et al., 2003. Leptin receptor polymorphism is associated with serum lipid levels and impairment of cholesterol lowering effect by simvastatin in Japanese men. Diabetes Res. Clin. Pract. 62, 169–175. http://doi.org/10.1016/S0168-8227(03)00163-3.
Tanofsky-Kraff, M., Han, J.C., Anandalingam, K., et al., 2009. The FTO gene rs9939609 obesity-risk allele and loss of control over eating. Am. J. Clin. Nutr. 90 (6), 1483–1488. https://doi.org/10.3945/ajcn.2009.28439.
The Allele Frequency Database https://alfred.med.yale.edu/alfred/index.asp, Accessed date: 24 January 2019.
The National Center for Biotechnology Information (NCBI). https://www.ncbi.nlm.nih.gov/, Accessed date: 15 April 2019.
Toperoff, G., Kark, J.D., Aran, D., et al., 2015. Premature aging of leukocyte DNA methylation is associated with type 2 diabetes prevalence. Clin. Epigenet. 7, 135–147. https://doi.org/10.1186/s13148-015-0069-1.
van der Vleuten, G.M., Kluijtmans, L.A., Hijmans, A., 2006. The Gln223Arg polymorphism in the leptin receptor is associated with familial combined hyperlipidemia. Int. J. Obes. 30 (6), 892–898. https://doi.org/10.1038/sj.ijo.0803234.

Walston, J., Silver, K., Bogardus, C., et al., 1995. Time of onset of non-insulin-dependent diabetes mellitus and genetic variation in the beta-3-adrenergic-receptor gene. N. Engl. J. Med. 333, 343–347. https://doi.org/10.1056/NEJM199508103330603.

Wauters, M., Considine, R.V., Chagnon, M., et al., 2002. Leptin levels, leptin receptor gene polymorphisms, and energy metabolism in women. Obes. Res. 10, 394–400. https://doi.org/10.1038/oby.2002.54.

World health organization Global action plan for the prevention and control of NCDs 2013-2020. Available at: http://apps.who.int/iris/bitstream/10665/94384/5/9789244506233_rus.Pdf, Accessed date: 24 March 2019.

Wu, Q., Saunders, R.A., Chin, K.V., 2010. The obesity-associated Fto gene is a transcriptional coactivator. J. Biochem. Biophys. Res. Commun. 401 (3), 390–405. https://doi.org/10.1016/j.bbrc.2010.09.064.

Yamakita, M., Ando, D., Tang, S., et al., 2010. The Trp64Arg polymorphism of the beta3-adrenergic receptor gene is associated with weight changes in obese Japanese men: a 4-year follow-up study. J. Physiol. Anthropol. 29 (4), 133–139. https://doi.org/10.2114/jpa2.29.133.

Yang, M., Xu, Y., Liang, L., et al., 2014. The effects of genetic variation in FTO rs9939609 on obesity and dietary preferences in Chinese han children and adolescents. PLoS One 9 (8), 154–165. https://doi.org/10.1371/journal.pone.0104574.

Yoshihara, A., Sugita, N., Iwasaki, M., et al., 2014. The interaction between beta-3 adrenergic receptor polymorphism and obesity to periodontal disease in community-dwelling elderly Japanese. J. Clin. Periodontol. 41 (5), 460–466. https://doi.org/10.1111/jcpe.12235.

Zavyalova, L.G., Denisova, D.V., Simonova, G.I., 2011. Association of polymorphism of the fto gene with excessive body weight in the Russian population. J. Siberian Sci. Med. 4, 5–11 (in Russian).

Zhang, Y., Proenca, R., Maffei, M., et al., 1994. Positional cloning of the mouse obese gene and its human homologue. Nature 372 (6505), 425–432. https://doi.org/10.1038/372425a0.

Zhu, L.Y., Hu, L.Y., Li, X.L., et al., 2010. Relationship between Trp64Arg mutation in the β3-adrenergic receptor gene and metabolic syndrome: a seven-year follow-up study. Chin. Med. J. (Engl) 123 (17), 2375–2378. https://doi.org/10.3760/cma.J.issn.0366-6999.2010.17.017.

Zyablitsev, S.V., Pankiv, V.I., Yuzvenko, T.Yu, 2016. Injecting the polymorphism rs1137101 of the leptin receptor gene into the development of tsukrovy diabetu type 2 and obesity in such diseases. J. Educ. Health Sport 6 (4), 503–514 (in Russian).

1000 Genomes project. http://www.internationalgenome.org/, Accessed date: 24 January 2019.