METABOLISM EFFECTOR LINKS IN DIET-INDUCED AND GENETICALLY-BASED OBESITY: A FULL-TRANSCRIPTOME STUDY OF LIVER TISSUE IN EXPERIMENTAL MODELS IN RODENTS

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ABSTRACT

Background. *When developing methods for personalized diet therapy of obesity, an urgent task is to study the molecular genetics features of the obesity pathogenesis using in vivo experimental models in laboratory animals.*
The aim. *To determine metabolism effector links in obesity based on a comparative analysis of full-transcriptome profiles of the liver tissue of mice and rats of various strains.*
Materials and methods. *We carried out a comparative analysis of the changes in liver transcriptome in rats and mice fed with diets of excessive energy value and exerting lipogenic effect. Data of full-transcriptome profiling using DNA microarray technology have been presented previously in 8 publications.*
Results. *In three strains of mice treated with a high-carbohydrate high-fat diet (HCHFD), a significant differential expression (DE) of 1849 genes was revealed, of which 74 genes responded jointly in at least two groups of animals. In Wistar and Zucker^{fa} rats, 2109 genes responded to the consumption of HCHFD, of which 242 genes responded jointly in two groups of animals. For rodents different in genetic predisposition to the development of diet-induced obesity, the groups of genes that responded with the opposite sign of DE (depending on the genotype) in reaction to the consumption of HCHFD were identified. Bioinformatical analysis allowed establishing the presence of 43 metabolic pathways, which are targeted for the applied experimental diets exposure, in rats and 77 pathways – in mice. Four of these pathways – the pathway of retinoid metabolism, PPAR signaling pathway associated with it the previous one, xenobiotics metabolism and drugs metabolism mediated by cytochrome P450 system – responded in all groups of animals (except for female mice). The importance of the expression of Tat gene encoding tyrosine aminotransferase in the modulation of biogenic amines synthesis in diet-induced obesity was shown, which may represent a new neurometabolic regulatory function of the liver in response to the consumption of high-calorie diets.*
Conclusion. *The analysis of the results of full-transcriptome studies showed that within each studied species (Rattus rattus and Mus domesticus) and animal sex, a number of genetic variants with a greater or lesser predisposition to the development of diet-induced obesity phenotype can be identified; and at the same time, within these variants, there is a largely similar pattern in the response of metabolism effector links to hypercaloric dietary intake. This pattern creates new prospects for translating the results of transcriptomic and metabolomic studies of laboratory animals into clinical practice in order to substantiate new approaches to personalized diet therapy of alimentary dependent diseases in patients with different genetic predisposition to obesity.*

Key words: *obesity, in vivo models, rats, mice, liver, transcriptome, metabolic pathways, neurometabolic function*

Received: 12.10.2022
Accepted: 25.05.2023
Published: 11.07.2023

For citation: Apryatin S.A., Trusov N.V., Gmoshinski I.V., Tutelyan V.A. Metabolism effector links in diet-induced and genetically-based obesity: A full-transcriptome study of liver tissue in experimental models in rodents. *Acta biomedica scientifica*. 2023; 8(3): 25-41. doi: 10.29413/ABS.2023-8.3.3

ЭФФЕКТОРНЫЕ ЗВЕНЬЯ МЕТАБОЛИЗМА ПРИ ДИЕТ-ИНДУЦИРОВАННОМ И ГЕНЕТИЧЕСКИ ДЕТЕРМИНИРОВАННОМ ОЖИРЕНИИ: ПОЛНОТРАНСКРИПТОМНОЕ ИССЛЕДОВАНИЕ ТКАНИ ПЕЧЕНИ НА ЭКСПЕРИМЕНТАЛЬНЫХ МОДЕЛЯХ У ГРЫЗУНОВ

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Статья поступила: 12.10.2022
Статья принята: 25.05.2023
Статья опубликована: 11.07.2023

РЕЗЮМЕ

Обоснование. *При разработке методов персонализированной диетотерапии ожирения актуальной задачей является изучение молекулярно-генетических особенностей его патогенеза с использованием экспериментальных моделей у лабораторных животных.*

Цель исследования. *Определение эффекторных звеньев метаболизма при ожирении на основе сравнительного анализа полнотранскриптомных профилей ткани печени мышей и крыс различных линий.*

Методы. *Проведён сравнительный анализ изменений транскриптома печени крыс и мышей, получавших рационы с избыточной калорийностью и липогенным действием. Данные полнотранскриптомного профилирования с использованием технологии ДНК-микрочипов были представлены ранее в 8 публикациях.*

Результаты. *У мышей трёх линий, получавших высокоуглеводный высокожировой рацион (ВУВЖР), выявлена достоверная дифференциальная экспрессия (ДЭ) 1849 генов, из которых 74 совместно ответили как минимум в двух группах животных. У крыс линий Wistar и Zucker^{fa} на потребление ВУВЖР ответили 2109 генов, из них 242 – в двух группах животных совместно. Для грызунов, различающихся по генетической предрасположенности к развитию диет-индуцированного ожирения, были определены группы генов, ответивших противоположной по знаку ДЭ (в зависимости от генотипа) в ответ на потребление ВУВЖР. Биоинформатический анализ позволил установить наличие у крыс 43, а у мышей – 77 метаболических путей, являющихся мишенями воздействия применяемых экспериментальных рационов. Из них 4 – путь обмена ретиноидов, сопряжённый с ним PPAR-сигнальный путь, метаболизм ксенобиотиков и метаболизм лекарственных препаратов под действием системы цитохрома P450 – ответили у всех групп животных (за исключением самок мышей). Показана важная роль экспрессии гена Tat, кодирующего тирозинаминотрансферазу, в модуляции синтеза биогенных аминов при диет-индуцированном ожирении, что, возможно, является новой нейрометаболической регуляторной функцией печени в ответ на потребление высококалорийных рационов.*

Заключение. *Анализ результатов полнотранскриптомных исследований показал, что в пределах каждого изученного вида (Rattus rattus и Mus domesticus) и пола животных можно выявить ряд генетических вариантов с большей или меньшей склонностью к развитию фенотипа диет-индуцированного ожирения; при этом в пределах каждого из этих вариантов отмечается во многом сходный характер ответа эффекторных звеньев метаболизма на потребление гиперкалорийного рациона. Эта закономерность создаёт новые перспективы для трансляции результатов транскриптомных и метаболомных исследований на лабораторных животных в клиническую практику для обоснования новых подходов к персонализированной диетотерапии алиментарно-зависимых заболеваний у пациентов, различающихся по генетической предрасположенности к ожирению.*

Ключевые слова: *ожирение, in vivo модели, крысы, мыши, печень, транскриптом, метаболические пути, нейрометаболическая функция*

Для цитирования: Апрятин С.А., Трусов Н.В., Гмошинский И.В., Тутельян В.А. Эффекторные звенья метаболизма при диет-индуцированном и генетически детерминированном ожирении: полнотранскриптомное исследование ткани печени на экспериментальных моделях у грызунов. *Acta biomedica scientifica*. 2023; 8(3): 25-41. doi: 10.29413/ABS.2023-8.3.3

INTRODUCTION

Alimentary dependent diseases, the leading development cause of which is an excess in energy value consumed and (or) unbalanced diet, represent one of the main challenges for modern medicine. In recent years, a large amount of experimental data has been accumulated that the pathogenesis of alimentary dependent diseases is based on the influence of excessive or unbalanced intake of macronutrients – fats and carbohydrates – on the expression of a large number of genes functionally related to the processes of lipogenesis, carbohydrate energy and protein metabolism, thermoregulation, systemic inflammation, circadian rhythms, and control of the amount of food consumed. The amplitude and trend of gene expression changes due to dietary factors are determined both by the genotype of the organism (the presence of polymorphisms in the genes of key enzymes, regulatory and transport proteins) and epigenetic factors (DNA methylation, histone acetylation, etc.), at the last in turn are also influenced by the composition of the diet [1]. The totality of issues of interaction between the genotype of an organism and such an important environmental factor as the composition of dietary nutrients in the formation of phenotype at the molecular, cellular, tissue, organ and organismic levels is the subject of study of a new section of nutrition science known as nutrigenomics [2].

The data obtained in the course of nutrigenomics research play an important role in the development of methods of personalized diet therapy of alimentary dependent diseases, taking into account such factors as the patient's genotype, nutritional status, stage and severity of the pathological process. Since nutrigenomics research in the clinic faces certain difficulties associated with the choice of a suitable biosubstrate, preclinical studies using *in vivo* experimental models of relevant diseases in laboratory animals characterized by different genetically-based propensity to disorders of lipid and carbohydrate-energy metabolism are in demand. A series of such models was developed based on the use of both genetically modified (mutant or knockout) and conventional strains of animals receiving excessive fat, simple sugars, or a combination of both (the so-called "Western diet" or the "cafeteria diet") [3]. Using up-to-date methods of molecular genetics and transcriptomics, a significant amount of data was obtained that in such animals during the development of obesity there are persistent changes in the expression of key gene groups responsible for metabolic processes in the liver and adipose tissue. For example, mice treated with a high-carbohydrate high-fat diet (HCHFD) for 1 year show differential expression (DE) of genes responsible for metabolic pathways of β-oxidation of fatty acids, steroid hormone biosynthesis and degradation, PPAR signaling, antigen processing and presentation, and proteasomal protein degradation [4]. Transcriptomics methods have been used to identify effector genes that are targets of dietary factors in dogs treated with a hypercaloric diet [5], in mice with diet-induced obesity (DIO) and non-alcoholic fatty liver disease [6]. Epigenetic factors contributing to the transmission of obesity phenotype from pregnant female rodents to their offspring have been characterized [7]. If suitable biomaterial (e. g., white adipose tissue specimens obtained during bariatric surgery) is available according to the requirements of medical ethics, it is possible to translate transcriptomic diagnostic criteria into clinical practice [8].

At the same time, the nature of nutrigenomic mechanisms determining a greater or lesser predisposition of the organism to the development of diet-induced obesity, which can be determined by comparative analysis of the results of transcriptomic studies performed using animals of different species and strains, is currently insufficiently covered in the literature. In a series of studies carried out in the Federal Research Center of Nutrition, Biotechnology and Food Safety during 2017–2022, the method of full-transcriptome analysis of liver tissue on a DNA microarray was used to identify effector links of metabolism – targets of the effect of minor bioactive substances of the diet (BAS), such as polyphenolic compounds, L-carnitine, amino acids, on models of DIO and spontaneous (genetically determined) obesity in rats and mice [9–13]. A number of rat strains (Wistar and Zucker^{fa}) and mice (C57Bl/6J, DBA/2J male strains, terahybrid mice) used in this study differed in the severity of obesity phenotype when consuming diets with excessive caloric intake, but the question of what differences in transcriptome parameters are the most significant in animals more or less prone to the development of DIO has not been sufficiently covered in these publications.

The aim of this work is to analyze data from transcriptomic studies on the nature of the liver transcriptome response to hypercaloric diets from a comparative perspective in rats and mice of different strains varying in susceptibility to the development of diet-induced or spontaneous obesity.

MATERIALS AND METHODS

The materials of full-transcriptome studies performed in mice and rats of different sexes and strains fed for 8–9 weeks a balanced control diet according to AIN-93M or modified semi-synthetic diets with a relative excess of fat (high-fat diet (HFD)), simple carbohydrate fructose (HFrD)), their combination (HCHFD) or cholesterol (high-cholesterol diet (HCD)) were used in this paper. Table 1 summarizes the experiments with references to publications revealing their design. All studies were conducted in compliance with the rules of biomedical ethics and were approved by the decision of the Ethics Committee of the Federal Research Center of Nutrition, Biotechnology and Food Safety (FSBIS) (record No. 4 of 20.04.2017).

After removal of animals from the experiment, liver tissue samples were obtained from them, from which total RNA was isolated using reagents of Agilent Total RNA Isolation Mini Kit (Agilent Technologies, Inc., USA) and full-transcriptome analysis was performed on Gene Expression Hy-

bridization Kit DNA microarrays (Agilent Technologies, Inc., USA) according to the Agilent One-Color Microarray-Based Gene Expression Analysis Low Input Quick Amp Labeling, version 6.8, protocol (Agilent Technologies, Inc., USA), using authorized certified equipment of the manufacturer (Agilent Technologies, Inc., USA).

The used DNA microarrays of different series contained between 30,000 and 32,000 annotated individual complementary sequences of the rat or mouse genome, including untranslated DNA sequences and splice variants. In all microarray experiments, 4 independent matrix RNA (mRNA) samples from each group of animals were analyzed. Microarrays were scanned on a SureScan Microarray Scanner (Agilent Technologies, Inc., USA). The DE value of genes determined by microarray analysis was expressed as the binary logarithm of the change in fluorescence (log_2FC) compared to control groups of animals treated with a balanced diet or to internal microarray controls (Spike-In). DE data were loaded into the «R» environment and bioinformatic analysis was performed with quantile normalization and further analysis in the limma package. The AnnotationDbi, org.Rn.eg.db, pathview, gage, and gageData packages were used to identify metabolic pathways using the Kyoto Encyclopedia of Genes and Genomes (KEGGs) international resource and visualize them. Statistical significance of expression changes was assessed using the the Benjamini – Hochberg multiple-correction T-test. Additionally, linear regression analysis was performed with calculation of Pearson correlation coefficients (PCC) between DE values (R) and their statistical significance (α). Annotation of genes that responded with statistically significant DE values during the deve-

TABLE 1

GROUPS OF ANIMALS USED IN EXPERIMENTS TO STUDY THE EFFECT OF OBESITY AND DYSLIPIDEMIA ON THE LIVER TRANSCRIPTOME

Species, sex of animals	Strain	*In vivo* model	Experimental diet composition[2]	References to publications
Female rats	Wistar[1]	Metabolic syndrome (MetS)	Semi-synthetic according to AIN-93M with replacement of drinking water with 30 % fructose solution (HFrD)	[10]
Female mice	C57Bl/6J[1]	MetS	HFrD	[9]
Female rats	Wistar[1]	Dyslipidemia (Dis)	Semi-synthetic according to AIN-93M with replacement of 0.5 % fat by weight of diet with cholesterol (HCD)	[10]
Female mice	C57Bl/6J[1]	Dis	HCD	[9]
Female rats	Wistar[1]	DIO	Semi-synthetic according to AIN-93M with replacement of 20 % of starch by weight of diet with fat (HFD)	[10]
Female mice	C57Bl/6J[1]	DIO	HFD	[9]
Male rats	Wistar[1]	DIO	Semi-synthetic according to AIN-93M with replacement of 20 % starch by weight of diet with fat, drinking water – with 20 % fructose solution (HCHFD)	[11, 12]
Male rats	Zucker[fa 3]	DIO	HCHFD	[13]
Female mice	C57Bl/6J[1]	DIO	HCHFD	[9]
Male mice	C57Bl/6J[1]	DIO	HCHFD	[13]
Male mice	DBA/2J[1]	DIO	HCHFD	[12, 13]
Male mice	DBCB[4] tetrahybrids	DIO	HCHFD	[13]

Note. [1] – obtained from the farm of Stolbovaya branch of the Scientific Center for Biomedical Technologies of FMBA of Russia; [2] – animals of control groups of each species, strain and sex received a balanced macronutrient and basic micronutrient semi-synthetic diet AIN-93M; [3] – obtained from Charles River farm (Italy); [4] – DBCB mice, hybrid of the 2nd generation (F2), were independently bred by the author of the study (S.A.A.) in the vivarium of the Federal Research Center of Nutrition, Biotechnology and Food Safety by sequential hybridization of mice of parental strains DBA/2J, CBA/lac (females) and BALB/c, C57Black/6J (males), as described in [13].

lopment of DIO in animals was performed using the international online resource Genemania (https://genemania.org) integrated with the PubMed (https://pubmed.ncbi.nlm.nih.gov) scientometric database. Set-theoretic analysis, construction of Venn diagrams and heat maps were performed using Venny 2.1 network application (https://bioinfogp.cnb.csic.es/tools/venny/) and MS Excel 2007 spreadsheets (Microsoft Corp., USA).

RESULTS AND DISCUSSION

Response of the liver transcriptome to the development of DIO in mice

As a result of full-transcriptome analysis of the livers of mice of four groups belonging to three strains (females and males of the C57Bl/6J strain, males of the DBA2J strain, males of the DBCB tetrahybrid strain), when comparing animals treated with HCHFD, with mice of control groups revealed DE at the level of more than 0.5 units modulo $\log_2$(FC) (i. e. 1.41-fold or more up or down) for 1,849 genes, of which 74 genes responded in at least two groups of animals together. Full data on the experiments performed have been presented previously [9, 13]. The analysis of the DE heat map of these genes (Fig. 1a) showed that the greatest similarity in the DE profile in response to the consumption of HCHFD is characterized by linear male C57Bl/6J and DBA2J mice, which, according to earlier studies, have a relatively high phenotypic resistance to the development of DIO [13], to which female C57Bl/6J mice adjoin, and DBCB tetrahybrid mice are characterized by the lowest degree of similarity with the other groups. Notably, it was the terahybrid mice that were the most phenotypically prone to the development of DIO according to the data of body weight gain and morphological signs of fatty liver disease (steatosis) [13]. Regression analysis showed that there was a statistically significant positive linear regression ($R = +0.562$; $\alpha = 0.005$) between the DE values of genes that responded jointly in C57Bl/6J and DBA2J males (Fig. 1b), whereas for DBA2J males and DBCB tetrahybrids (Fig. 1c), in contrast, the corresponding regression was negative ($R = -0.689$; $\alpha < 0.001$), and for DE comparisons between DBCB mice and C57Bl/6J males and between males and females of the latter strain (Fig. 1d, e), there was no statistically significant regression ($\alpha > 0.1$).

Thus, the genetic expression profile in different strains of mice phenotypically similar in resistance to developing of DIO is largely consistent, whereas in mice that differ markedly in their propensity to develop DIO (such as DBA2J and DBCB), the DE trend appears to be largely opposite.

Annotation of genes that responded with DE in mice of different strains showed several groups of genes that responded with opposite sign values of DE to HCHFD intake in DBCB tetrahybrid mice and DBA/2J, C57Bl/6J linear animals, which are among the parental strains of the indicated tetrahybrid. Of these, the most representative were two groups of genes belonging to metabolic pathways of lipid metabolism, the first of which included *Tff3, Scd1, Pltp* (positive DE in DBCB and negative DE in DBA2J and/or C57Bl/6J mice), and the second group included *Pparg, Usp18, Crot, Ifi202b, Mvk, Sqle, Msmo1, Idi1* (the DE trend was opposite to the first group of genes). *Tff3* expression in rodent liver is highly susceptible to changes in models of early diabetes and fatty liver disease. TFF3 protein reduced hepatic steatosis induced by a high-fat diet by increasing PPARα-mediated fatty acid oxidation [14]. The function of *Scd1* is to synthesize oleate from stearate and palmitooleate from palmitate. Suppression of *Scd1* expression protects mice from developing hepatic steatosis and obesity [13]. The *Pltp* gene is responsible for the synthesis of phospholipids, which are part of lacrimal fluid, pulmonary surfactant, and others. Cholesterol, phospholipid, apolipoprotein A1 and apolipoprotein B, and high-density lipoprotein concentrations in plasma were decreased in mice with knockout of this gene. Besides, PLTP protected mice from the development of atherosclerosis without causing lipid accumulation in the liver [15].

Pparg encodes a nuclear transcription factor that affects genome elements called peroxisome proliferators and which are responsible for regulating a complex set of genes involved in metabolism and peroxisomal β-oxidation of fatty acids, adipocyte differentiation, and glucose homeostasis. Its expression is associated with other *Ppar* family genes as well as *Fabp4*, *Mapk1*, etc. PPARγ functions in an ensemble with retinoid X receptors (RXR). The role of *Pparg* in the activation of Ito fat-storing cells in the liver has been proven [16]. The *Crot* gene is responsible for the attachment of medium-chain fatty acid residues to carnitine during their peroxisomal β-oxidation [13]. *Usp18* stimulates lipolysis, fatty acid oxidation in transformed lung epithelial cells, and protects mice from hepatic steatosis and the development of insulin resistance [17]. The expression of the gene *Ifi202b*, which is involved in the differentiation of adipose stem cells into mature adipocytes, is closely associated with it. Increased expression of *Ifi202b* stimulates adipogenesis in mice and humans [18]. A similar expression profile in tetrahybrid mice compared to linear mice was observed in a functionally closely related group of genes *Msmo1, Mvk, Idi1*, and *Sqle1* involved in cholesterol and steroid metabolism. Of these, *Msmo1*, as reported in the literature, has the ability to suppress adipogenesis and differentiation of immature adipocytes [19]. On the other hand, *Sqle* has been identified as one of the genes that contribute to obesity in mice [20].

A group of genes involved in mineral metabolism responded differently to DE values in linear and tetrahybrid mice. Of these, *Mt1* and *Mt2* were characterized by negative DE in DBA/2J mice and positive DE in DBCB mice. The proteins encoded by these genes are metallothioneins, which play a vital role in the homeostasis of a number of essential (copper, zinc, manganese) and toxic (cadmium, lead, mercury) trace elements; the amount of synthesized metallothioneins is associated with the development of obesity and hepatic steatosis [13]. In connection with the revealed facts, it is appropriate to note that an earlier study on DBA/2J and DBCB mice treated with HCHFD revealed a sig-

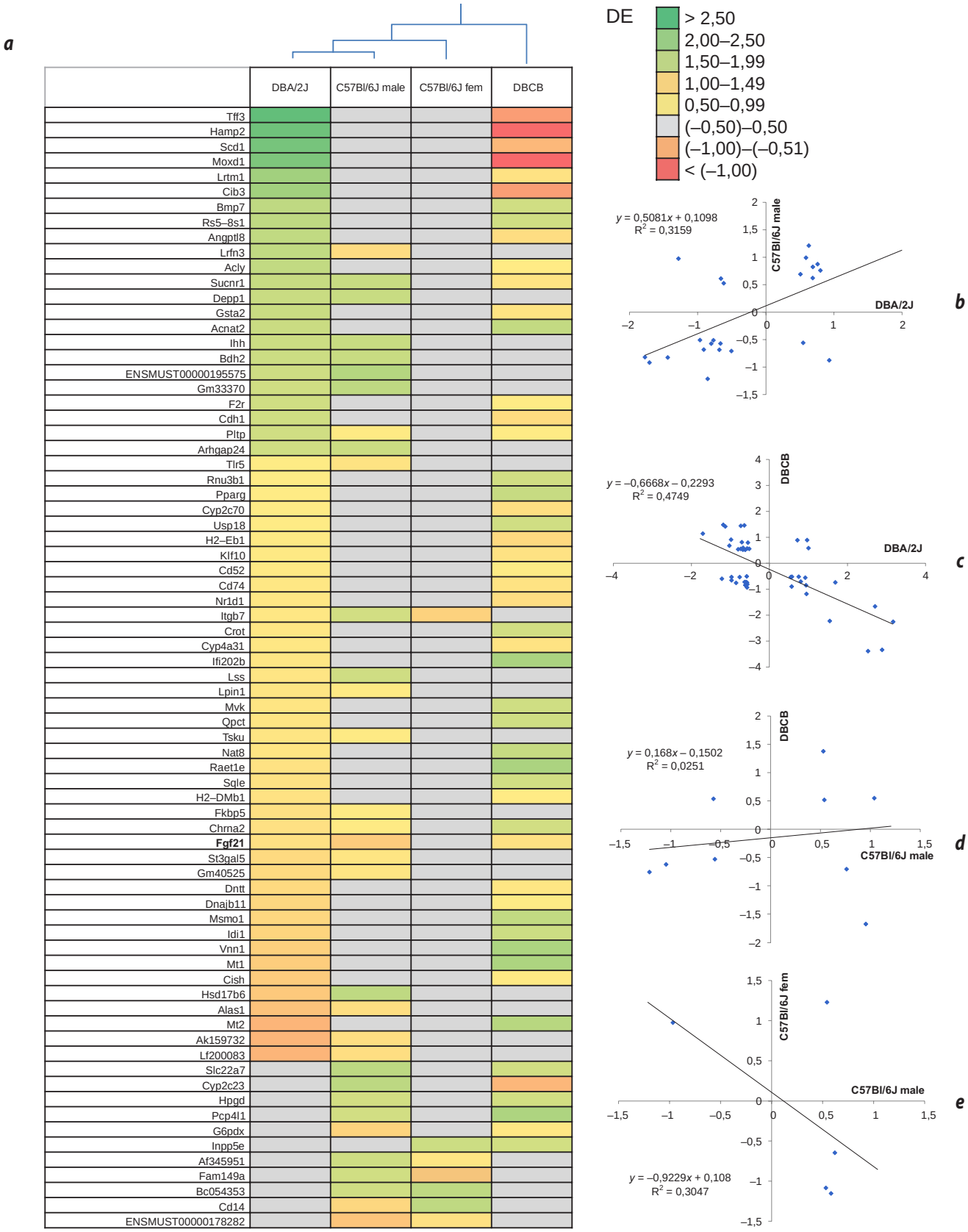

FIG. 1.

Comparative analysis of the differential expression values of genes that jointly responded to the consumption of high-carbohydrate high-fat diet in DBA/2J mice (males), C57Bl/6J (males and females), and tetrahybrid DBCB (males): ***a*** *– heat map of differential expression of genes in mice of 4 groups;* ***b*** *– DBA/2J – C57Bl/6J (males) regression;* ***c*** *– DBA/2J – DBCB regression;* ***d*** *– C57Bl/6J (males) – DBCB regression;* ***e*** *– C57Bl/6J (males) – C57Bl/6J (females) regression. The degree of similarity (X) was calculated according to the formula: X = N × (C – M), where N is the total number of jointly responding genes; C is the number of genes with the same sign of differential expression; M is the number of genes with the opposite sign of differential expression*

nificant effect of their genotype on the bioaccumulation indices of a number of divalent cations of trace elements (lead, manganese, copper, cadmium, etc.) [13].

The opposite profile of DE (positive in linear mice and negative in DBCB tetrahybrids) was characteristic of *Moxd1* and *Hamp2*. *Moxd1* encodes a copper binding protein that has dopamine β-monooxygenase activity and is involved in the biosynthesis of the biogenic amines octopamine and norepinephrine. The expression values of *Moxd1 and Dbh* (dopamine beta hydroxylase) are closely related to each other as analyzed by https://genemania.org. The significance of these data will be further discussed below. The product of *Hamp2* expression is the peptide hormone hepcidin, which regulates iron metabolism. Elevated plasma levels of this protein have been observed in obese children, which correlates with reduced iron status and the development of systemic inflammation as assessed by interleukin (IL) 6 production [21].

A number of genes involved in antigen presentation, cytokine signaling pathways, and inflammation development also responded with the opposite sign of DE to HCHFD consumption in DBCB tetrahybrid and linear mice. These include the *Sucnr1, Cdh1, Raet1e, and F2r* genes. The specific DE profiles of these genes may be correlated with differences in cytokine production in HCHFD-receiving DBCB tetrahybrids and linear DBA/2J mice in response to the consumption of a number of BAS [13].

Response of the liver transcriptome to the DIO development in rats

When analyzing the transcriptome of male Zucker^{fa}, female Wistar, and male Wistar rats (two repeats of the experiment) treated with HCHFD, the DE was detected for 2,109 genes at the level modulo more than 0.5 $\log_2$(FC) units compared to the group of animals of the same strains receiving the control diet. Of these, 174 genes responded jointly in males of the Zucker^{fa} and Wistar strains; 11 in males and females of the Wistar strain; 6 in males of the Zucker^{fa} and females of the Wistar strains; and 51 in males of the Wistar strain in two repeated experiments. Details of the liver transcriptome obtained in these experiments have been reported previously [10, 11, 13].

A heat map of the distribution of genes according to the value of their DE (Fig. 2a) shows that the greatest similarity in the DE profile is observed for males of the Wistar and Zucker^{fa} strains, whose difference from Wistar females is more significant. According to Figure 2b, there is a statistically significant (R = +0.422; α < 0.001), although not very powerful positive regression between DE values for Wistar and Zucker^{fa} males. The data in Figure 2c show that the regression between the DE of the genes reproduced in two repeats of the experiment on male Wistar rats is positive, highly statistically significant (R = +0.947; α < 0.001), and the regression line runs near the origin. For males and females of the Wistar strain (Fig. 2d), the regression relationship of the DE of jointly responded genes is statistically significant and negative (R = –0.651; α = 0.042), that is, the response of the transcriptome to HCHFD consumption is to a certain extent discordant between them.

Annotation of genes that responded with opposite sign of DE to HCHFD consumption in male Zucker^{fa} and Wistar rats identified several groups of genes responding during DIO development, including *Abhd2, Cpt1a, Kiss1, Myc, Myc, Prlr, Ppp1r3c, Tsc22d1*, and *Upp2*. *Cpt1a,* encoding carnitine palmitoyltransferase I, is characterized by negative expression in Zucker^{fa} males and positive expression in Wistar males. Positive expression of this gene, responsible for the process of β-oxidation of fatty acids, is considered as a normal response of the organism to the consumption of excess fat [13]; it can be assumed that this mechanism is disturbed in Zucker^{fa} rats.

A similar DE profile is characteristic of the *Kiss1* gene encoding the precursor neuropeptide kisspeptin, which is presumed to have an anorectic action (reducing appetite and food intake) [13]. According to numerous data in the literature, decreased *Kiss1* expression is observed during the development of diet-induced obesity [13]. The same trend in rats of the two strains has the expression of *Abdh2* (hormone-sensitive lipase subunit), *Upp2* (cofactor of hepatic X receptor, PPARα- and HNF-4α-signaling pathways), *Tsc22d1* (coregulator of PPARα), *Prlr* (prolactin receptor, which stimulates lipid oxidation and thermogenesis in brown adipose tissue, one of the cofactors of dopamine action [22]) and *Ppp1r3c* (a protein phosphatase responsible for the regulation of glycogen storage in liver cells).

The opposite trend, i. e. positive in Zucker^{fa} rats and negative in Wistar males, of DE was observed for the *Myc* gene. It encodes a multifunctional nuclear phosphoprotein that plays a role in cell cycle, apoptosis and malignant transformation. Its functions include triggering the development of liver fibrosis and regulation of glucose-responsive genes through the CHREBP signaling pathway [23].

The next group of genes that contrastingly responded to HCHFD consumption in rats of the two strains under study includes genes responsible for implementing the function of proto-oncogenes (i. e., intracellular protein phosphorylation signaling cascade) and stimulation of apoptosis. In addition to the analyzed *Myc*, these include *Jun, Atf3, Dusp6,7, Epcam*, and *Casp4*. In particular, Jun protein kinase, positively expressed in Zucker^{fa} and negatively expressed in Wistar, is a component of the JNK signaling chain responsible for the developmental consequences of oxidative stress, including apoptosis, and insulin resistance. According to [24], *Jun* expression is upregulated in mice with hepatic steatosis induced by HFD consumption. *Jun* is also functionally related to *Mlxipl*, which regulates glucose metabolism, and *Kiss1*, discussed above.

Among the differentially expressed genes involved in inflammation and cytokine signaling pathways, *Ccl3* (Mip-1a), *CD274,276* and *Cish* genes are characterized by positive DE in Zucker^{fa} and negative DE in Wistar, while *Ackr2, Bcl6* and *Bmf* genes have the opposite DE profile. Of these, *Bcl6* is a functionally relevant gene whose expression product suppresses IL-6-stimulated macrophage proliferation [25], IL-18 synthesis, and Th2 cell differentiation, which together may contribute to the control of systemic inflammation induced by excessive adipogenesis. In Zucker^{fa} rats, this molecular mechanism appears to be disrupted.

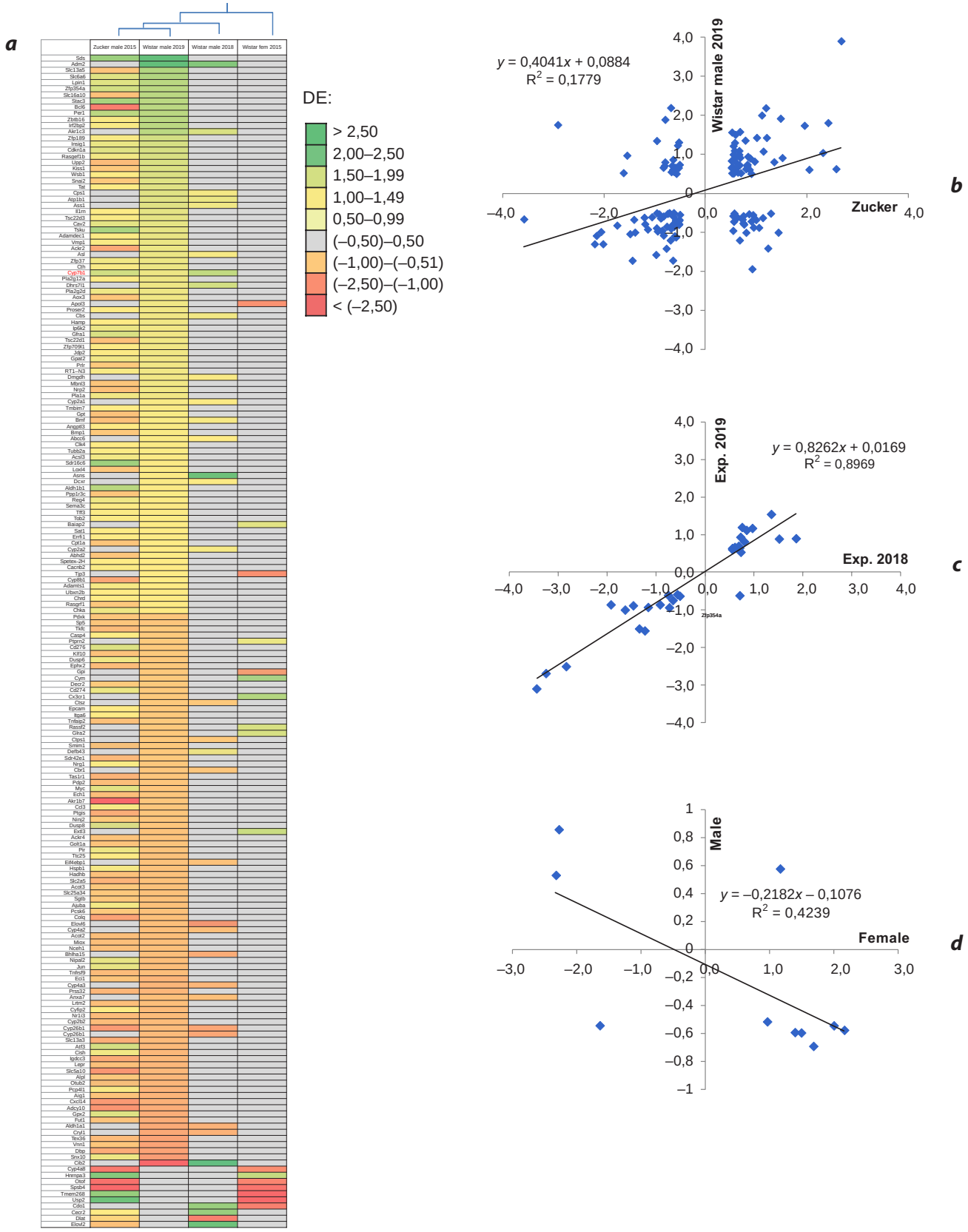

FIG. 2.

Comparative analysis of the differential expression values of genes that jointly responded to the consumption of high-carbohydrate high-fat diet in the Zucker^{fa} rat lines (males), Wistar rats (males (2 experiments) and females): ***a*** *– heat map of differential expression of genes in 4 groups of rats;* ***b*** *– Zucker^{fa} – Wistar (males, experiment of 2019) regression;* ***c*** *– regression for two groups of Wistar males in experiments of 2018 and 2019;* ***d*** *– Wistar females and males groups regression. The degree of similarity (X) was calculated according to the formula: X = N × (C – M), where N is the total number of jointly responding genes; C is the number of genes with the same sign of differential expression; M is the number of genes with the opposite sign of differential expression*

It is also necessary to point out the opposite trend in Zucker^{fa} and Wistar rats of the HCHFD-induced DE of a number of genes (*Aox3, Bmp1, Gpx2, Hspb1, Lox4, Pir*) responsible for the processes of mineral metabolism (binding of iron, copper), the function of bioantioxidants (selenium) and inhibition of oxidative stress. The same applies to genes of the steroid hormone metabolic pathway such as *Cyp8b1* and *Nrg1*, as well as amino acid metabolism and transport (*Gpt, Slc16a10*). At the same time, alanine aminotransferase (ALT), whose expression is decreased in Zucker^{fa} rats and activated in Wistar rats upon HCHFD consumption, is a gene suppressed by insulin and, like *Tat* discussed below, stimulated by glucocorticoids [26]. ALT plays an important role in protein catabolism and gluconeogenesis [13].

Comparing features of the liver transcriptome response to HCHFD between rats and mice

Genes in the PPARγ signaling pathway, xenobiotic metabolism by cytochromes P450, retinol metabolism, transamination, and other pathways of aromatic amino acid metabolism were common with respect to differential gene expression for both rodent species' metabolic regulatory processes.

Typically, genes related to transamination and apoptosis were activated in both rodent species.

The processes common to the two rodent species indicate that transamination and apoptosis are among the most relevant processes in the regulation of metabolic reactions in rodents in response to the consumption of a high-carbohydrate high-fat diet (HCHFD), determining the ratio of catabolic to anabolic reactions along with other transaminases such as aspartate aminotransferase (AST) and ALT. The differences between the two rodent species may be due to differences in ATP-dependent processes, regulatory cascades (PPARγ), mechanisms of energy metabolism (coenzyme A), oxidative stress (glutathione) and apoptosis between species and animal lines.

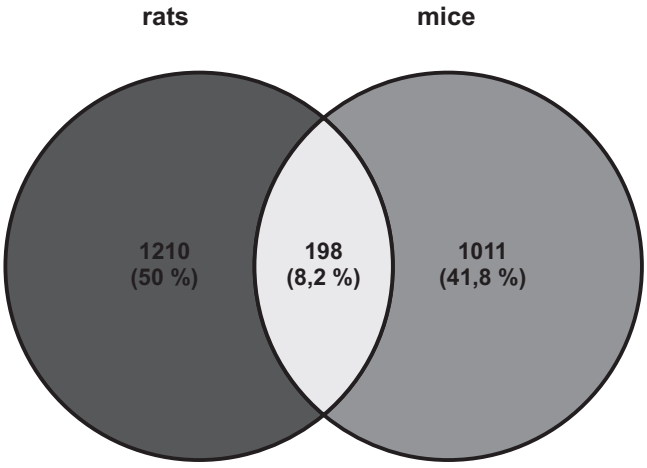

FIG. 3.
Venn diagram of differential expressions of genes in rats and mice of different strains that jointly responded to the consumption of high-carbohydrate high-fat diet

Comparative analysis of the results of studies of different mouse and rat strains revealed 198 DE genes common to both rodent species (Fig. 3). The genes that jointly responded by differential expression to HCHFD consumption in at least one of the studied mouse strains and in rats were *Cish, Lpin1, Nat8, Pcp4l1, Tsku, Bmp7, Cd52, Cd74, Depp1, Fgf21, Idi1, Ihh, Klf10, Lss, Nr1d1, Pparg, Tff3, Usp18, Vnn1*, with the first five of this list responding in both Zucker^{fa} rats and Wistar males. Some of the genes listed above play a key role in the regulation of lipid oxidation, lipogenesis, and adipose tissue proliferation.

For example, *Lpin1* encodes phospholipase C, which generates diacylglycerol, a coregulator of a large number of transcription factors. Overexpression of *Lpin1* in mice suppresses the development of alcoholic hepatitis by inhibiting lipolysis and reducing the amount of fatty acids entering the liver, and suppression of the activity of this gene is observed in the development of metabolic syndrome [13]. *Tsku* expression is associated with *Ppara* and is increased in non-alcohol related steatohepatitis [27]. *Fgf21* is an important hepatokine with pleiotropic function known as a metabolic regulator of glucose and lipid homeostasis with anorexigenic effects [28]. Finally, *Vnn1,* considered as a biomarker of toxic kidney injury and under the control of PPARα, plays a role in inflammation, oxidative stress and cell proliferation [29].

DE of the *Tat* gene encoding tyrosine aminotransferase was detected under HCHFD exposure in male Wistar and Zucker^{fa} rats, male DBA/2J mice and female C57Bl/6J mice, and spontaneously obese db/db mice compared with their parental C57Bl/6J strain (described in detail below). Other differentially expressed genes common to both rodent species were *Plekhf1* (pleckstrin homology domain), *Atp1b1* (Na^+/K^+-ATPase b1-subunit), and *Chka* (choline kinase alpha). The Na^+/K^+-ATPase b1-subunit is a plasma membrane pump with multiple physiological functions. It maintains ion homeostasis, which is crucial for cell survival, differentiation and apoptosis [30]. A similar pattern of positive differential expression in liver tissue to the *Tat* gene was obtained for the *Plekhf1* (*LAPF*) gene in the Wistar rat strain and the DBA/2J inbred mouse strain, but not C57Black/6J. The function of this gene in the pathogenesis of obesity is currently unclear. Chka is the second ATP-dependent protein to show directly opposite hepatic expression patterns in mice and rats. This may be due to differences in the rate of ATP-dependent energy metabolism processes between the two rodent species (it is higher in mice).

The role of genes such as *Idi1, Cish, Pparg*, *Tff3* and *Usp18* in adipogenesis and development of DIO has been discussed above.

Metabolic pathways of rats and mice of different strains responding to consumption of hypercaloric and hyperlipidemic diets

Bioinformatic analysis of DE genes identified metabolic pathways (KEGGs) statistically significantly altered by experimental hypercaloric diets. Details of the findings on KEGGs targets of different dietary exposures

in experiment are presented in previous publications [9–13].

In mice of the C57Bl/6J strains (male and female), DBA/2J males (in two repeats of the experiment) and DBCB tetrahybrids, the effect of HCHFD on 77 metabolic pathways was detected at the $p < 0.05$ level of statistical significance. Heat map analysis of these influences, plotted against the statistical significance of the effect value (p_{val}), showed (Fig. 4a) that male C57Bl/6J and DBA/2J mice and, on the other hand, DBA/2J and DBCB tetrahybrids form two similar clusters, and their difference from male C57Bl/6J mice appears to be more significant. Thus, mouse sex appears to be a stronger determinant of the trend of changes in KEGGs during DIO compared to genotype (strain). Venn diagram set-theoretic analysis (Fig. 4b) indicates that there were no metabolic pathways that responded simultaneously to HCHFD in mice of all sexes and strains. However, in male C57Bl/6J and DBA/2J mice, 9 metabolic pathways responded jointly to HCHFD consumption, 9 metabolic pathways also responded in DBA/2J and DBCB tetrahybrids, 5 in male C57Bl/6J and DBCB mice, and 1 in female and male C57Bl/6J mice. Four metabolic pathways (mmu00982 Drug metabolism – cytochrome P450; mmu00980 Metabolism of xenobiotics by cytochrome P450; mmu00830 Retinol metabolism; mmu00330 Arginine and proline metabolism) responded in all groups of mice except for C57Bl/6J females.

Noteworthy differences in the nature of the effects of HCHFD on the mmu00830 Retinol metabolism and associated mmu03320 PPAR signaling pathways in male DBA/2J and DBCB mice contrastingly differ in the development of the DIO phenotype in response to HCHFD consumption. Figure 4c shows that DBCB tetrahybrids have a partially bypassed metabolic block at the stage of conversion of vitamin A to its active all-trans retinal form by retinol dehydrogenase (RDH), which may lead to reduced production of 9-cis-retinoate. The latter, in turn, is a ligand for the RXR receptor of the PPAR signaling pathway (Fig. 4d), differentially expressed in mice of the two strains. Along with the presence of differential expression of PPARβδ receptors, this leads to a series of multidirectional changes (partial or complete metabolic blocks) in the processes of transport and β-oxidation of fatty acids, which is probably reflected in the differences in the phenotype of these animals discussed above.

In male Zucker^{fa} and Wistar rats in two replications of the study (the experimental data are presented in detail in publications [11, 13]), the intake of HCHFD caused a total of changes in 43 metabolic pathways, of which 7 were common to all conducted studies (Fig. 5a). When comparing two parallel tests on male Wistar rats [11, 13], 18 out of 39 (46 % of the total) identified KEGGs were observed to match, which is a fairly good indicator for the reproducibility of transcriptomic studies according to the MAQC Consortium (cited in [13]) in light of the fact that the studies were performed on different cohorts of animals and using DNA microarrays of different series.

All 7 metabolic pathways, including rno00830 Retinol metabolism, rno00980 Metabolism of xenobiotics by cytochrome P450, rno00982 Drug metabolism – cytochrome P450, rno03320 PPAR signaling pathway, rno00590 Arachidonic acid metabolism, rno00140 Steroid hormone biosynthesis, rno01040 Biosynthesis of unsaturated fatty acids, that responded to HCHFD consumption in all groups of rats responded simultaneously to this exposure in at least part of the mouse groups, with the first four of these KEGGs responding simultaneously in three of the four mouse groups studied (all males). This indicates a fairly high measure of confidence in the identification of these KEGGs as targets of the effects of hypercaloric diet in various models of DIO in rodents. In total, 31 metabolic pathways responded jointly in rats and mice in at least two experiments to HCHFD consumption (Fig. 5b).

Using the heat map method (Fig. 5c), the measure of similarity in the response of different metabolic pathways to different hypercaloric (HFD, HFrD, HCHFD) and hyperlipidemic (HCD) diets in rats and mice was analyzed to identify possible dietary, genotypic, and gender patterns in this. The results of clustering by animal groups are shown in Figure 5d in the form of a graph ("phylogenetic tree"). They show a clear separation of female C57Bl/6J mice from all other animal groups (both rats and mice). On the other hand, two other clusters are evident in the response to the investigated diets, of which one is represented by male C57Bl/6J and DBCB mice on HCHFD and female Wistar rats on HFD and the other by male DBA/2J mice on HCHFD and female Wistar rats on HFrD and HCD. Female Wistar rats and male Zucker^{fa} rats, the most prone to develop DIO, do not fall into any of these clusters, demonstrating high specificity of their metabolic response.

When comparing the features of metabolic pathways in male Wistar and Zucker^{fa} rats contrasting in the severity of the obesity phenotype, it was noted that in the rno00830 Retinol metabolism pathway in Wistar rats treated with HCHFD, there is a partially overcoming metabolic block in the enzymatic pathways for the formation of all-trans-retinoate and the thermodynamically irreversible formation of 9-cis-retinoate from it, whereas in Zucker^{fa} rats both of these metabolic blocks apparently cannot be overcome. On the other hand, a metabolic block of glucuronidation of all-trans-retinoate occurs in Zucker^{fa} rats, which can lead to inhibition of its clearance. Taken together, these effects could hypothetically affect the ratio of cis- and trans-isomers of retinoic acid, which play different roles in the regulation of intracellular processes through interaction with RXR receptors (see above for a mouse example). However, proof of the possibility of such a mechanism is impossible without kinetic modeling of the corresponding enzymatic reactions, for which there is currently insufficient experimental data.

The rno00590 Arachidonic acid metabolism pathway may also play a significant role in the difference in response to HCHFD consumption between Zucker^{fa} and Wistar rats. Namely, Wistar rats receiving HCHFD have a metabolic block (which cannot be overcome or partially overcome) in the biosynthesis of PGF2α, 5-HETE and 15(S)-HETE, in contrast to Zucker^{fa} rats in which these reactions are, in contrast, activated. The consequence of these differences may

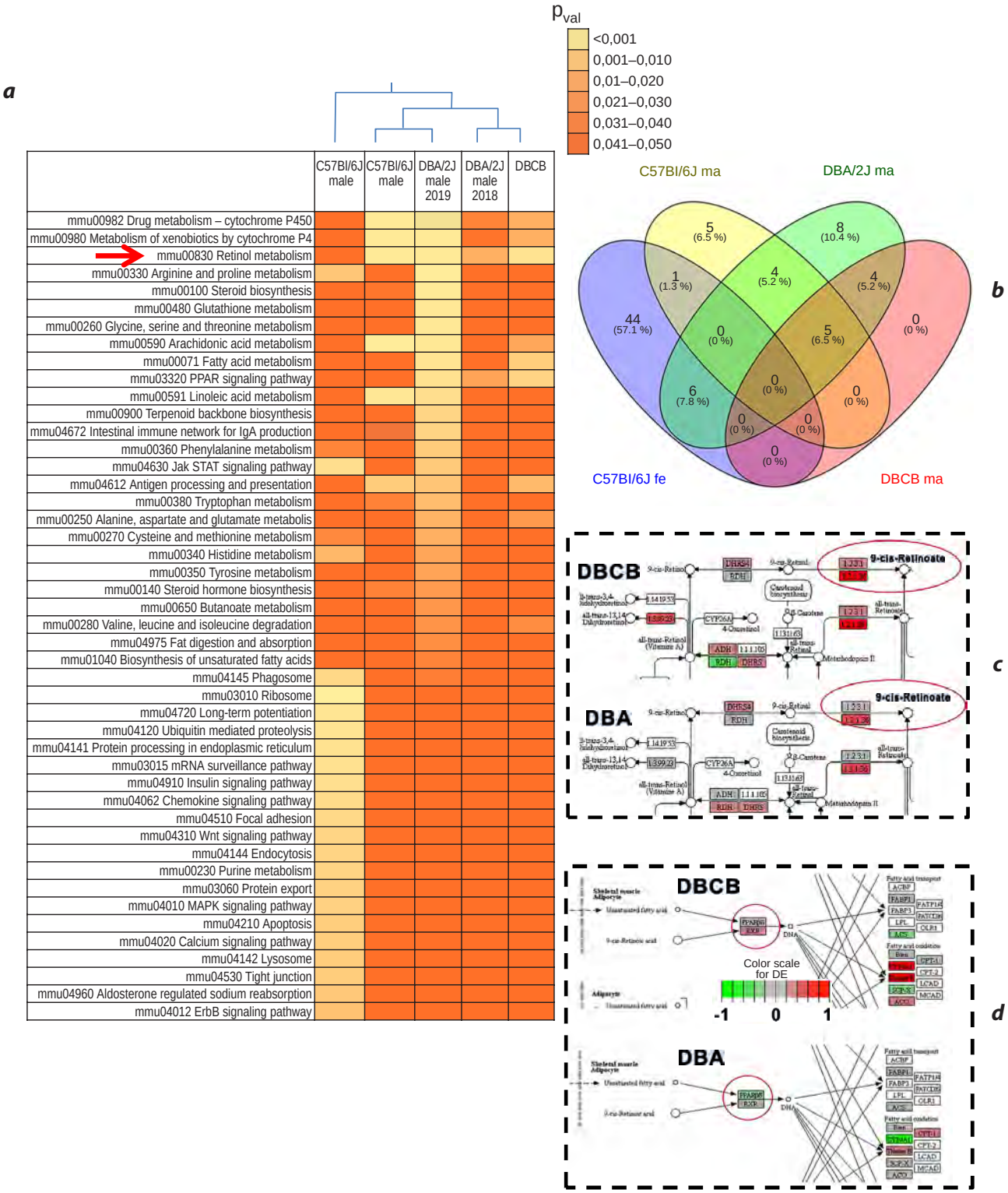

FIG. 4.

Comparative analysis of metabolic pathways (KEGGS) significantly ($p_{val} < 0.05$) responding to the consumption of high-carbohydrate high-fat diet in C57Bl/6J mice (males and females), DBA/2J (males, 2 experiments) and DBCB tetrahybrids: ***a*** *– heat map of the p_{val} values of the metabolic pathways in 5 groups of mice;* ***b*** *– Venn diagram of the distribution of metabolic pathways that jointly responded in different groups of mice;* ***c*** *– fragments of the metabolic pathway mmu00830 Retinol metabolism in DBCB and DBA/2J mice;* ***d*** *– fragments of the metabolic pathway mmu03320 PPAR signaling pathway in DBCB and DBA/2J mice. C57Bl/6J fe – C57Bl/6J females; C57Bl/6J ma – C57Bl/6J males; DBA/2J ma – DBA/2J males; DBCB ma – DBCB males. The measure of similarity was calculated by the criterion of the number of significantly ($p < 0.05$) jointly responded KEGGS*

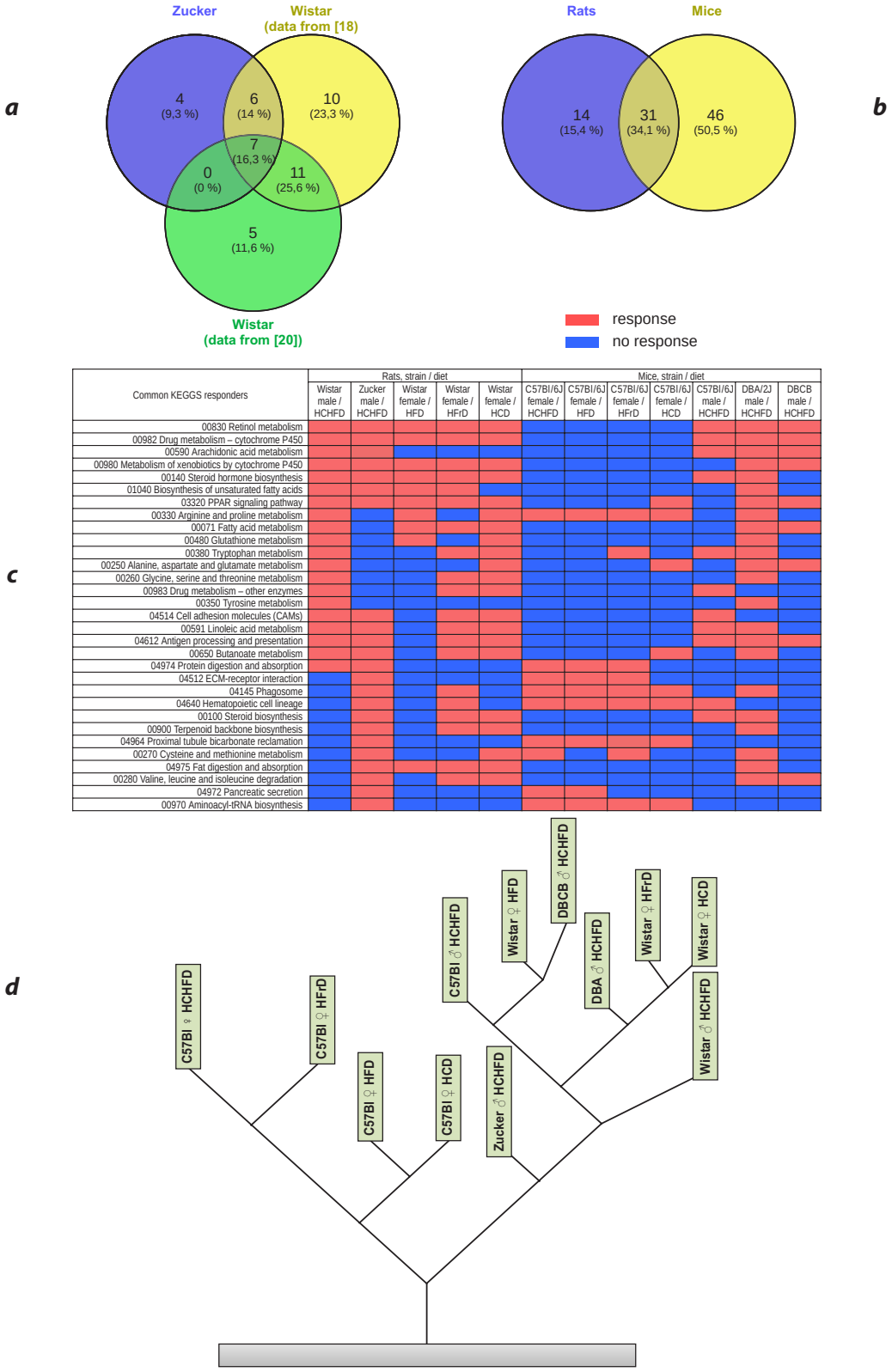

FIG. 5.

Comparative analysis of metabolic pathways (KEGGS) that jointly responded in rats and mice of different sexes and strains to the consumption of high-carbohydrate high-fat diet, as well as to the consumption of high-fructose, high-fat and high-carbohydrate diets (the last three – only in female C57Bl/6J mice and female Wistar rats): ***a*** *– Venn diagram of the distribution of metabolic pathways that responded to the consumption of high-carbohydrate high-fat diet by the groups of rats;* ***b*** *– Venn diagram of the distribution of the number of metabolic pathways that jointly responded to the consumption of high-carbohydrate high-fat diet in rats and mice (all studied sexes and lines);* ***c*** *– heat map of the responses of 31 metabolic pathways which jointly responded in rats and mice fed with various types of hyperlipidemic and hypercaloric diets;* ***d*** *– graph demonstrating a measure of similarity in the KEGGS response profile in different strains of rats and mice fed with the experimental diets*

be the different nature of the response in rats of these two strains of the oxylipin profile – derivatives of polyunsaturated fatty acids, which play an essential role in the expression regulation of a large number of genes, including those involved in the processes of lipogenesis, lipid (fat) metabolism, immune response and inflammation [13].

A significant limitation of the approach used in this paper is the incomplete coverage of diet-induced transcriptomic changes in mice and rats of both sexes and different strains, which is determined by the totality of data presented in the original studies [9–13] under comparable conditions. However, even on such a fragmentary sample of combinations of species, sex, and strains of animals, it became possible to reveal a general pattern consisting in the fact that under the influence of feeding hypercaloric diets in both rats and mice, the same groups of genes that respond oppositely with DE values to one and the same dietary factor, depending on the degree of genetic predisposition to the DIO development, are allocated. With a certain degree of schematic characterization, they may include 1) genes involved in the lipogenesis regulation and lipid metabolism; 2) genes of signaling pathways of proto-oncogenes and intracellular messengers, including those responsible for cell differentiation and apoptosis; 3) genes of inflammation factors, regulatory molecules of immune cells, cytokines and their receptors; 4) genes of proteins involved in the binding, transport and biological function of trace elements; 5) enzyme genes of amino acid metabolism, especially those capable of controlling the availability of substrates for the synthesis of biogenic amines (including trace amines and classical neurotransmitters) involved in the regulation of metabolic rate, physical mobility, eating behavior and appetite, which will be further discussed below in relation to the *Tat* gene.

It is particularly important to note a pattern common to both rodent species, consisting of the effect of an energy-abundant diet on the expression of genes involved in the regulation of metabolism via the PPARγ signaling pathway, xenobiotic metabolism by cytochromes P450, retinol metabolism, amino acid metabolism (including TAT transamination), and regulation of apoptosis. In contrast, the remaining differentially expressed genes were characterized not only by interspecies differences between mice and rats, but also by interstrain differences within mice of the two inbred strains and the tetrahybrid.

Role of *Tat* gene expression in metabolic effects of DIO in rat and mouse models

The *Tat* gene encodes the tyrosine aminotransferase enzyme (EC 2.6.1.5), which catalyzes the reversible reaction of transferring the amino group from the amino acid tyrosine to α-ketoglutarate to form *p*-hydroxyphenylpyruvate and glutamate, respectively. The TAT enzyme, like other aminotransferases, is vitamin B_6-dependent and plays an important role in the metabolic pathway of tyrosine biotransformation. The enzyme can also utilize phenylalanine as an amino group donor and phenylpyruvate as its acceptor [31]. In fact, through a metabolic link catalyzed by hepatic TAT, there is regulation of the amount of the conditionally essential amino acid tyrosine available for the synthesis of the biogenic amine dopamine and its derivatives, including adrenaline, epinephrine, and norepinephrine. A genetic defect in *Tat* in human's results in tyrosinemia type 2 accompanied by profound neurological disorders.

As follows from the data of Table 2, a significant DE of the *Tat* gene was detected under the influence of HCHFD in male Wistar and Zucker^{fa} rats, male DBA/2J mice and female C57Bl/6J mice, as well as in spontaneously obese db/db mice compared to their parental strain C57Bl/6J (the experimental data are presented in the monograph [13]). In all cases, except for db/db mice and C57Bl/6J females, factors leading to the development of obesity induced a positive DE of *Tat*. Interestingly, the differential expression rate of the *Tat* gene was more than twofold higher for Wistar rats ($\log_2$(FC) = 1.212; p_{val} = 0.002; adj.p_{val} = 0.067) compared to the control diet, while it was only 46 % higher for rats of the Zucker^{fa} strain ($\log_2$(FC) = 0.553; p_{val} = 0.041; adj.p_{val} > 0.1). Moreover, *Tat* expression was reduced almost twofold in db/db mice ($\log_2$(FC) = –0.962; p_{val} = 0.001; adj.p_{val} = 0.006).

The role of transamination reactions and tyrosine metabolism as effector links of metabolism in DIO and obesity caused by genetic factors is indicated by changes in AST activity and De Ritis ratio (AST/ALT) values in blood plasma of not only homozygous (*in vivo* model of hyperactivity accompanied by increased resistance to DIO development) but also heterozygous DAT-KO rats [13]. These results indicate the effect of excessive amounts of extracellular dopamine in the synaptic cleft of dopaminergic neurons in the striatum on the regulation of metabolic processes through activation of catabolic processes, including lipid and carbohydrate metabolism. As a consequence, the DAT-KO knockout rats receiving HCHFD showed a decrease in key parameters of the obesity phenotype (body weight, relative liver weight, white retroperitoneal adipose tissue, etc.) in comparison with «wild-type» animals, which were the parental Wistar strain for DAT-KO rats.

As in the case of *DAT* gene knockout, diet-induced increases in TAT expression and activity can reduce extracellular dopamine levels in synapses of dopaminergic neurons, acting in particular on the nigrostriatal and mesolimbic dopamine systems of the brain. The observed changes in TAT expression, along with ALT and AST transaminases, may affect the severity and trend of catabolic and/or anabolic organism reactions, the features of eating behavior and the ability to control the amount of food consumed.

Consistent with these findings are the results of a transcriptomic study indicating that both rats and mice more prone to develop an obesity phenotype (Zucker^{fa} rats, db/db mice, female C57Bl/6J mice) show less *Tat* expression compared with animals phenotypically relatively more resistant to developing obesity (male Wistar rats, DBA/2J mice). This includes results showing the presence of DE in mice of different strains of *Moxd1 and Dbh* genes responsible for some steps in the metabolism of dopamine and its derivatives. The expression of the *Hamp2* (hepcidin) gene discussed above may apparently be associated with dopa-

mine metabolism through the role of this gene in the regulation of tissue iron status, which is an important cofactor in a number of stages of metabolism of this biogenic amine.

Based on these findings, it can be concluded that there is a TAT-dependent effect on the activation of transamination processes in the liver by altering the rate of catabolic reactions, as well as regulation of dopamine levels via tyrosine utilization via the non-dopamine pathway, possibly representing a novel neurometabolic regulatory function of the liver in response to the consumption of high-calorie diets.

Analysis of intergenic interactions using the "genetic networks" tool implemented in Genemania resource shows the relationship of *Tat* expression with other aminotransferases, including Gpt (ALT), *Got* (AST), as well as *Vnn1, Fos, Jun* and some other genes involved in lipogenesis and response to HCHFD. This indicates a legitimate role for *Tat* as a metabolic link in the response of the body phenotype to the consumption of a calorie-abundant diet. *Tat* expression increases in mice receiving hypercaloric diet in the setting of metabolic correction by ingestion of *Luffa cylindrica* dietary fiber, and, according to the authors, the mechanism of this effect is mediated by a favorable effect of intestinal microflora metabolites on gene expression in the host liver [32]. In our study on Wistar rats, the content of TAT protein was increased in the liver cells of rats treated with HCHFD by immunohistochemical analysis [33].

Another putative mechanism of metabolic regulation of *Tat* expression is attributed to the systemic effects of glucocorticoids. Thus, *Tat* is known to be stimulated by corticosterone administration or by immobilization stress accompanied by massive glucocorticoid release [34]. In this regard, it is relevant to analyze the relationship between *Tat* expression and metabolic pathways (KEGGs) of steroid hormone biosynthesis and metabolism, which, according to transcriptomic studies, are the targets of the effects of HCHFD and other hypercaloric diets in different rodent species and strains.

CONCLUSION

Thus, the use of the method of full-transcriptome profiling made it possible to reveal on models of diet-induced obesity, hyperlipidemia and metabolic syndrome in rats and mice the nutrigenomic effects associated with the different character of the influence of excessive consumption of dietary fat and (or) carbohydrates on the transcriptome of liver tissue in animals more or less hereditarily predisposed to the development of DIO or hereditarily determined obesity, steatosis and dyslipidemia. A significant role of *Tat* gene expression, encoding tyrosine aminotransferase, in the regulation of metabolic reactions in rodents in response to HCHFD consumption and in the development of obesity was shown.

The results obtained in a large number of cases are reproducible, coincide in animals of different species and strains characterized by similar phenotype, and agree with the data of analysis of integral, biochemical, traceelement and morphological parameters. At the same time, within each studied species (*Rattus rattus* and *Mus domesticus*) and a certain sex of animals, it is possible to identify a number of genetic variants with a greater or lesser propensity to develop the DIO phenotype; in addition, within these variants, a largely similar trend of the transcriptome response to dietary influence is noted.

This summarizing result allows one to remember the scientific position of the outstanding geneticist Nikolai I. Vavilov, expressed in the 1st half of the XX cen-

TABLE 2

DIFFERENTIAL EXPRESSION OF THE *Tat* GENE IN DIFFERENT GROUPS OF ANIMALS IN RESPONSE TO THE DEVELOPMENT OF SPONTANEOUS OR DIET-INDUCED OBESITY AND DYSLIPIDEMIA

Models	Species, sex of animals	Strain	Log(FC)	p_{val}	adj. p_{val}	References to publications
MetS (HFrD feeding)	Female mice	C57Bl/6J	–0.554	< 0.001	0.08	[9]
DIO (HCHFD feeding)	Female mice	C57Bl/6J	0.775	< 0.001	**0.043**	[9]
Dyslipidemia (HCD feeding)	Female mice	C57Bl/6J	0.403	0.002	–	[9]
Spontaneous obesity (balanced diet)	Male mice	db/db[1]	–0.962	0.001	**0.006**	[13]
DIO (HCHFD feeding)	Same	DBA/2J	1.449	< 0.001	**0.004**	[12]
DIO (HCHFD feeding)	Male rats	Zucker	0.553	0.041	–	[13]
DIO (HCHFD feeding)	Male rats	Same	1.212	0.002	0.067	[13]

Note. [1] – mouse strain with leptin receptor gene knockout. Mice of the C57Bl/6J strain, which is the "parental" strain for this knockout strain [13], receiving the same diet served as a control in this experiment.

tury, that "Species and genera that are genetically close are characterized by similar series of hereditary variability with such accuracy that, knowing a number of forms within one species, one can foresee the finding of parallel forms in other species and genera". This postulate, known as the Law of homologous series in hereditary variability, was empirically derived from studies on genetic variants of bread cereal plants. However, N.I. Vavilov himself noted that the «law of homologous series» can also be applied to animals. If the assumption that the different responses of the transcriptome and its associated metabolome to the consumption of energy-dense food are similar across species (including humans), this improves the prospects for translating results obtained from *in vivo* experiments into clinical practice.

The general trend of the transcriptome response and its derived metabolome may contribute to both the development of DIO and the formation of resistance to it by increasing energy expenditure and controlling the amount of food consumed. Which of these variants is most likely to be realized depends on the genotype of the organism, i. e., the presence of allelic polymorphisms of key "obesity genes". The search for such genes is still a crucial task of nutrigenetics, but it is obvious from the available literature data that not all problems in this field have been solved. In particular, there are insufficient data on gene expression products that could be targets of planned personalized dietary interventions. Of the genes examined in this paper, the most numerous clinical evidences of association with the pathogenesis of obesity are known for polymorphisms of the *Pparg* gene. At the same time, there is reason to believe that the scope of work on the search for «candidate obesity genes» can be significantly narrowed, and these studies can be further structured and targeted to take into account genes that are part of metabolic pathways that respond contrastingly depending on the obesity phenotype. In accordance with the data presented in this paper, it is reasonable to search for such genes as part of the metabolic pathways of retinoid metabolism, RRAR signaling, steroid hormone metabolism, oxylipins (including eicosanoids), trace elements, aromatic amino acids, which are precursors of trace amines and neurotransmitters with known neuroregulatory function.

Source of funding

The research was carried out at the expense of the subsidy for fulfillment of the state task within the framework of the Program of Fundamental Scientific Research (topic of the Ministry of Education and Science of Russia No. 0529-2015-0006 "Search for new molecular markers of alimentary dependent diseases: genomic and post-genomic analysis") and with the support of the grant of the Russian Science Foundation No. 17-16-01043 "Search for effector links of metabolism regulated by dietary factors in obesity for the development of innovative specialized food products".

Conflict of interest

The authors of this article confirmed that there is no conflict of interest to be reported.

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