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Lipolysis is accompanied by immune microenvironment remodeling in adipose tissue of obesity with different exercise intensity

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Abstract. – OBJECTIVE: Obesity and overweight are risk factors for chronic disease worldwide. The purpose of this study was to compare the transcriptome of exercise-induced fat mobilization in obese people, and to explore the effect of different exercise intensity on the correlation of immune microenvironment remodeling and lipolysis in adipose tissue.

MATERIALS AND METHODS: Microarray datasets of adipose tissue before and after exercise were downloaded from the Gene Expression Omnibus. Then, we used gene-enrichment analysis and PPI-network construction to elucidate the function and enrichment pathways of the differentially expressed genes (DEGs) and to identify the central genes. A network of protein-protein interactions was obtained using STRING and visualized with Cytoscape.

RESULTS: A total of 929 DEGs were identified between 40 pre-exercise (BX) samples and 65 post-exercise (AX) samples from GSE58559, GSE116801, and GSE43471. Among these DEGs, adipose tissue-expressed genes were duly recognized. Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) enrichment analyses indicated that DEGs were mostly enriched in lipid metabolism. Studies have found that mitogen-activated protein kinase (MAPK) signaling pathway and forkhead box O (FOXO) signaling pathway are up-regulated, while Ribosome, coronavirus disease (COVID-19) and IGF-1 gene are down-regulated. Although we found the up-regulated genes that noted IL-1 among others, and the down-regulated gene was IL-34. The increase of inflammatory factors leads to changes in cellular immune microenvironment, and high-intensity exercise leads to increased expression of inflammatory factors in adipose tissue, leading to inflammatory responses.

CONCLUSIONS: Exercise at different intensities leads to the degradation of adipose and is accompanied by changes in the immune microenvironment within adipose tissue. High intensity exercise can cause the imbalance of immune microenvironment of adipose tissue while causing fat degradation. Therefore, moderate intensity and below exercise is the best way for the general population to reduce fat and weight.

Key Words:

Exercise, Adipose, Bioinformatics analysis, MAPK-signaling pathway, IGF-1, IL, Immune microenvironment.

Introduction

During the past 30 years, the rates of obesity and overweight have increased dramatically for all age groups, and they are likely to continue unabated into the future[1,2]. Although China currently exhibits the highest number of obese individuals of any country[3,4], obesity constitutes an increasingly serious public-health concern globally[5,6]. An increase in obesity can result in serious diseases such as cardiovascular disease, diabetes, osteoarthritis, and hyperlipidemia[7]. Adipose tissue affects insulin sensitivity, blood pressure, endothelial function, fibrinolytic activity, inflammatory response, and many important pathophysiological processes[8]. It has also been shown that irisin, regarded as a messenger of the muscle-fat-bone-brain axis, plays an important role in promoting the Browning of white adipose tissue[9].

The most common means used to lose weight include medication, diet, exercise, and surgery. Many previous studies[10-14] have shown that aerobic exercise produces potential benefits in weight loss, and studies[15] have indicated that it can induce genomic changes in adipose tissue that may affect adipose metabolism.

Obesity leads to changes in neutrophil, monocyte and lymphocyte counts as well as reduced T- and B-cell-induced proliferation, resulting in a decrease in the body's immune function[16]. Macrophages are the most abundant immune cells in the body, and in obese individuals, the shift from

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the M2 anti-stress state to the M1 pro-inflammatory state in adipose tissue is accelerated, producing insulin resistance[17]. It has been shown that regular physical activity has many benefits for the body, reducing the incidence of disease and decreasing inflammation; high-intensity exercise can lead to tissue damage and trigger local and systemic inflammatory responses[18]. Scholars[19] have shown that both acute and chronic exercise can have an impact on the immune system. Resistance exercise increases the phagocytic activity of neutrophils[20]. And prolonged but not strenuous exercise may increase the protective function of the immune system[21]. Therefore, we speculate that the intensity of exercise degrades fat while causing a remodeling of the immune microenvironment in adipose tissue.

Although exercise induces the mobilization of adipose, the underlying mechanism of action remains to be elucidated, and bioinformatics analyses of the effects of different exercise intensity on the correlation of immune microenvironment remodeling and lipolysis in adipose tissue have not yet been executed. In this research, therefore, we investigated the regulation of exercise training on gene expression, and we used Kyoto Encyclopedia of Genes and Genomes (KEGG) and Gene Ontology (GO) bioinformatics analysis to identify changes between immune microenvironment remodeling and adipose degradation in adipose tissue.

Materials and Methods

Acquisition of Microarray Data

Microarray data of adipose tissue before and after exercise were obtained using the GEO database. Our filter criteria were the availability of (1) Homo sapiens expression profiling by arrays, (2) adipose tissue before and after exercise from combined adipose biopsies, (3) samples that consisted of three datasets, (4) datasets that contained integral information regarding the samples (5) one biopsy sample per subject for analysis (no replicates). And finally selected three datasets GSE58559, GSE116801, and GSE43471 as the test set, including 40 pre-exercise (BX) samples and 65 post-exercise (AX) samples (Table I).

Normalizing Data and Identifying Differential Genes

The primitive files downloaded from the GEO database were preprocessed and normalized using the robust multiarray average (RMA) method on the https://www.aclbi.com/static/index.html#/geo site. The gene analysis of differences between samples was performed using the Limma package (Bioconductor, Roswell Park Comprehensive Cancer Center, NY, USA), and after the *p*-value was obtained, multiple hypothesis testing and correction were performed. The *p*-values for differentially expressed genes (DEGs) were determined by controlling for the false discovery rate (FDR), and the corrected *p*-value was adjusted[22]. The threshold *p*-value for the DEGs was determined by controlling the FDR, and the *p*-value was adjusted[23]. The screening criteria were $log_2FC=4$ and a *p*-value <0.01. In addition, we performed differential gene analysis on the samples before and after exercise to look for inflammatory factors ($log_2FC=2$, p-value<0.05).

Visualize the DEGs

To better visualize the DEGs, we used the https://www.aclbi.com/static/index.html#/geo to create heatmaps and volcano maps. At the same time, we used the heatmap pack (Bioconductor, Roswell Park Comprehensive Cancer Center, NY, USA) to generate a heatmap.

Gene Pathway and Functions Analysis

The distribution trends of predefined groups of genes in the gene table were assessed using GSEA (available at: http://www.gsea-msigdb.org/gsea/index.jsp) to determine their contribution to the phenotype. We downloaded GSEA_4.1.0 and the c5: GO gene set (available at: c5.all.v7.1.symbols.gmt) for functional enrichment analysis. The https://www.aclbi.com/static/index.html#/geo site was then used to analyze the GO enrichment of the DEGs, and a chord diagram was created to visualize these rich results. KOBAS 3.0 (available at: http://kobas.cbi.pku.edu.cn/kobas3)[24] is an online database widely used for gene/protein pathway enrichment and functional annotation, and it was used for the KEGG-pathway and reacted enrichment analyses of the DEGs. The significantly enriched pathways and functions were selected using $p<0.01$ and $log_2FC=4$.

Construction and Visualization of The Protein-Protein Interaction (PPI) Network

Use the STRING database (Stockholm, Swiss) to build a protein-protein interaction (PPI) network of our identified DEGs, a precomputed database in which associations between proteins are assigned based on high-throughput experiments, gene fusions, co-occurrence, literature mining, co-expression analysis, and computational pre-

Table I. Information on the selected microarray datasets.

GEO accession	Platform	Samples BX, AX	Sex	Source tissue	Exercise regimen
GSE58559	GPL10558	0 25	------	Adipose	16 weeks, intense exercise ------
GSE116801	GPL570	10 10	Male	Adipose	12 weeks, cycling 60-80 minutes/day, five days/week
GSE43471	GPL6947	30 30	Female	Adipose	24 weeks, moderate intensity 45 minutes/day, five days/week

dictions. Interactions were visualized with Cytoscape v. 3.8.2 (available at: http://www.cytoscape.org/)[24]. The Molecular Complex Deletion (MCODE) plugin was used to identify gene cluster modules[25]. In this network, according to the filter criteria, four modules were identified, cluster 1 (score: 34.914, 36 nodes and 1,222 edges), followed by cluster 2 (score: 9, 9 nodes and 72 edges), cluster 3 (score: 4.8, 6 nodes and 24 edges), and cluster 4 (score: 4.429, 15 nodes and 42 edges) (Figure 3).

Network Module Analysis

The network modules were then analyzed using the MCODE plugin in Cytoscape[26].

Statistical Analysis

SPSS 26.0 (IBM Corp., Armonk, NY, USA) was used for general statistical analysis. GraphPad Prism 8.0 software (La Jolla, CA, USA) and R Studio (Boston, MA, USA) were used to generate plots. Spearman's correlation analysis was performed to calculate correlation coefficients. Data were presented as the mean ± SD values. $p<0.05$ commonly indicated a statistically significant difference.

Results

Identification of GSE58559, GSE116801, and GSE43471 DEGs

Finally, we selected the datasets GSE58559, GSE116801, and GSE43471 – which included 40 BX samples and 65 AX samples – to analyze and identify the DEGs. Comparing genes in the BX and AX samples, a total of 929 DEGs were identified, comprising 93 down-regulated and 836 up-regulated genes ($\log_2$FC=4, $p<0.01$). We visualized and identified differential genes with volcano plot and heatmap analysis (Figure 1 and Figure 2). Table II describes the 20 most significantly up-regulated and down-regulated genes, with the majority of significantly down-regulated genes recognized as ribosomal genes.

KEGG Pathway and GO Analyses

Functional and pathway enrichment analysis was performed using the GSEA software (available at: http://www.gsea-msigdb.org/gsea/index.jsp), clusterProfiler package (Bioconductor, Roswell Park Comprehensive Cancer Center, NY, USA) and the online tool KOBAS 3.0. First, the expression profiles of all genes in BX and AX samples were uploaded to GSEA and then performed an overall-level GO-enrichment analysis of the expression profiles using the c5: GO gene set; the screening criteria for significant gene sets were designated as $p<0.01$ and log2FC=4. Next, the https://www.aclbi.com/static/index.html#/geo site was ultimately used to perform KEGG- and GO-pathway analyses on 929 DEGs, including 836 up-regulated and 93 down-regulated genes. In the KEGG-pathway analysis, up-regulated pathways were enriched in cyclic adenosine monophosphate (cAMP) signaling pathway, mitogen-activated protein kinase (MAPK) signaling pathway, forkhead box O (FOXO) signaling pathway ubiquitin-mediated proteolysis, neuroactive ligand-receptor interaction, among other pathways (Figure 1C); while down-regulated pathways were enriched in ribosome, coronavirus disease (COVID-19), and lysosome, among others (Figure 1D). In the GO analysis, up-regulated pathways were enriched in regulation of lipid metabolic process, regulation of small molecule metabolic process, glycerolipid metabolic process, and alcohol metabolic process, among others (Figure 1E); while down-regulated pathways were enriched in mRNA catabolic process, translational initiation, establishment of protein localization to membrane, and RNA catabolic process, in addition to others (Figure 1F).

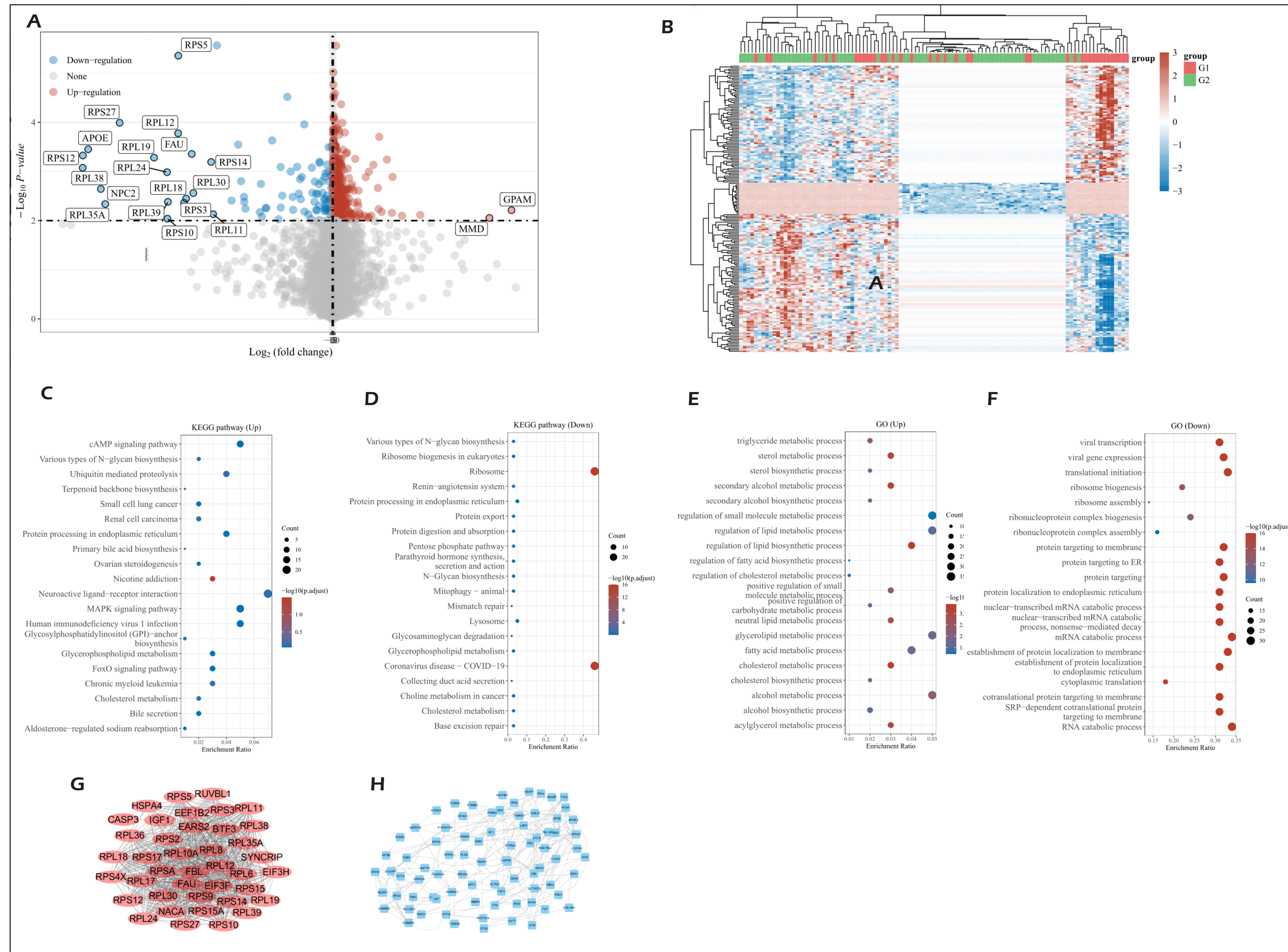

Figure 1. Volcano maps, heat maps, and up-regulated and down-regulated gene maps. **A**, Heatmap of DEGs between the BX and AX samples. Red rectangles represent high expression, and blue rectangles represent low expression. **B**, Volcano plot of DEGs between the BX and AX samples. The red plots represent up-regulated genes, the black plots represent unaltered genes, and the blue plots represent down-regulated genes. **C,** Top 20 upregulated KEGG pathways. **D,** Top 20 down-regulated KEGG pathways. **E,** Top 20 upregulated GO pathways. **F,** Top 20 downregulated GO pathways. **G,** Network analysis with ≥30 gene links. **H,** Network analysis with 10-30 gene links.

Table II. Top 20 upregulated and downregulated differentially expressed genes.

Upregulated expressed genes			Downregulated expressed genes		
Gene	Log_2FC	adj. *p*-value	Gene	Log_2FC	adj. *p*-value
GPAM	780.56	0.14	*RPL38*	-1,091.82	0.10
MMD	684.23	0.15	*RPS12*	-1,091.12	0.10
CES1	381.96	0.14	*APOE*	-1,067.77	0.10
MRAP	302.72	0.15	*NPC2*	-1,012.40	0.11
CAP2	261.45	0.10	*RPL35A*	-993.07	0.13
TNS3	258.52	0.14	*RPS27*	-930.33	0.10
PMEPA1	236.70	0.10	*RPL19*	-780.05	0.10
ATP2B4	213.74	0.14	*RPL24*	-723.47	0.10
TPST2	203.08	0.10	*RPS10*	-721.64	0.16
PTPN11	200.94	0.13	*RPL39*	-719.70	0.12
CCDC83	197.28	0.11	*RPL12*	-675.17	0.10
SLC29A4	177.31	0.10	*RPS5*	-674.22	0.02
GMFB	168.44	0.15	*RPS3*	-649.01	0.12
GASK1B	158.27	0.10	*RPL18*	-640.10	0.12
FOXO1	156.76	0.15	*FAU*	-615.60	0.10
WFS1	155.53	0.15	*RPL30*	-608.79	0.11
CDKN2B	144.00	0.10	*RPS14*	-530.86	0.10
ZNF595	141.30	0.15	*RPL11*	-522.00	0.14
HSD17B4	123.10	0.15	*RPS9*	-515.50	0.11
KRT40	119.15	0.14	*RPL6*	-505.61	0.02

PPI Network Analysis and Module Identification

Analysis of 929 DEG by STRING and visualization using the Cytoscape plugin MCODE revealed a total of 5,580 interactions involving three modules. When the plug-in was used to search for genes with more than 30 connections to other genes, a total of 40 genes were found (Table III), of which 6 were upregulated and 34 were downregulated. The BioGPS database (available at: http://biogps.org/#goto=welcome) is utilized to annotate DEGs[27]. Most of the down-regulated genes were ribosomal genes while *IGF-1* was among the upregulated genes. *IGF-1* principally exists in blood, and its primary functions include lowering blood glucose and lipids and promoting cellular differentiation. Thus, a network of ≥30 gene links containing 40 genes is constructed (Figure 1G), and a network with having connections between 10 and 30 to other genes that included 98 genes (Figure 1H).

Inflammatory Cytokines

Interleukins are lymphoid factors that interact with leukocytes or immune cells and belong to the families of cytokines that serve as blood cell growth factors. They coordinate and interact with one another to collectively complete hematopoiesis and immune-regulatory functions. Interleukins also play an important role in transmitting messages, inflammatory responses and regulating activated immune cells. When GEO was performed on differential gene expression analyses to find genes associated with interleukin as a blood inflammatory factor, a total of 18 genes were found, 17 of which were up-regulated and one gene that was down-regulated. Then, it was noted that up-regulated genes included *IL-1*, *IL-2, IL-3, IL-5, IL-17* and *IL-36*, and down-regulated genes included *IL-34* (Table IV). According to the analysis of the samples in the GSE43471 dataset before and after exercise, *IL-17* was significantly up-regulated, and *IL-34* was significantly down-regulated in the differential genes. Analysis of samples in the GSE116801 dataset before and after exercise showed that no inflammatory factors were found in the differential genes obtained ($p<0.05$, $log_2FC=2$).

Discussion

Obesity and overweight are risk factors for cardiovascular disease, diabetes, chronic kidney disease, numerous types of cancer, and a range of musculoskeletal diseases. With changes in lifestyle and diet, the prevalence rates of overweight or obesity have increased considerably worldwide[28]. Physical activity can cause the Browning of white adipose tissue[29], improve the sensitivi-

Table III. Use of GSE58559, GSE43471, and GSE116801 datasets, and identification of 40 hub genes using three algorithms of cytoHubba.

Gene symbol	Description	log_2FC	adj. p-value	Regulation
SYNCRIP	glycerol-3-phosphate acyltransferase, mitochondrial	39.15	0.13	Up
IGF1	insulin like growth factor 1	30.82	0.15	Up
HSPA4	heat shock protein family A (Hsp70) member 4	13.11	0.68	Up
CASP3	caspase 3	11.00	0.13	Up
EARS2	glutamyl-tRNA synthetase 2, mitochondrial	10.76	0.10	Up
RUVBL1	RuvB like AAA ATPase 1	9.51	0.12	Up
FBL	Fibrillarin	-57.66	0.10	Down
EEF1B2	eukaryotic translation elongation factor 1 beta 2	-123.26	0.14	Down
RPL36	ribosomal protein L36	-134.94	0.10	Down
EIF3H	eukaryotic translation initiation factor 3 subunit H	-139.46	0.12	Down
EIF3F	eukaryotic translation initiation factor 3 subunit F	-183.57	0.10	Down
RPS15	ribosomal protein S15	-220.21	0.12	Down
RPSA	ribosomal protein SA	-236.55	0.16	Down
RPL8	ribosomal protein L8	-291.31	0.13	Down
BTF3	basic transcription factor 3	-313.82	0.11	Down
NACA	nascent polypeptide associated complex subunit alpha	-380.92	0.13	Down
RPS15A	ribosomal protein S15a	-386.16	0.12	Down
RPS2	ribosomal protein S2	-387.22	0.17	Down
RPL17	ribosomal protein L17	-388.34	0.10	Down
RPL10A	ribosomal protein L10a	-399.94	0.11	Down
RPS4X	ribosomal protein S4 X-linked	-441.77	0.12	Down
RPS17	ribosomal protein S17	-466.11	0.11	Down
RPL6	ribosomal protein L6	-505.61	0.02	Down
RPS9	ribosomal protein S9	-515.47	0.11	Down
RPL11	ribosomal protein L11	-521.96	0.14	Down
RPS14	ribosomal protein S14	-530.86	0.10	Down
RPL30	ribosomal protein L30	-608.79	0.11	Down
FAU	FAU ubiquitin like and ribosomal protein S30 fusion	-615.60	0.101	Down
RPL18	ribosomal protein L18	-640.10	0.12	Down
RPS3	ribosomal protein S3	-649.01	0.12	Down
RPS5	ribosomal protein S5	-674.22	0.02	Down
RPL12	ribosomal protein L12	-675.17	0.10	Down
RPL39	ribosomal protein L39	-719.70	0.12	Down
RPS10	ribosomal protein S10	-721.64	0.16	Down
RPL24	ribosomal protein L24	-723.47	0.10	Down
RPL19	ribosomal protein L19	-780.05	0.10	Down
RPS27	ribosomal protein S27	-930.33	0.10	Down
RPL35A	ribosomal protein L35A	-993.07	0.13	Down
RPS12	ribosomal protein S12	-1,091.12	0.10	Down
RPL38	ribosomal protein L38	-1,091.82	0.10	Down

ty of tissue to insulin, promote fat oxidation and regulate lipid metabolism[30-33]. In our study, then herein we analyzed the GSE116801, GSE58559, and GSE43471 databases for exploring the effect of different exercise intensity on the correlation of immune microenvironment remodeling and lipolysis in adipose tissue.

Our study found that MAPK signaling pathway expression was significantly up-regulated after exercise. Activation of this signaling pathway is involved in energy metabolism associated with inflammation[34] and can regulate insulin sensitivity, the pathogenesis of obesity and diabetes[35-38], and promote white adipose tissue

Table IV. Expression of the interleukin blood-inflammatory factors.

IL	Regulation
IL36A	Up
IL36B	Up
IL17RE	Up
IL23R	Up
IL5	Up
IL12B	Up
IL3	Up
IL17A	Up
IL36RN	Up
IL9R	Up
IL22RA2	Up
IL17C	Up
ILDR2	Up
ILDR1	Up
IL2	Up
IL1RAP	Up
IL17B	Up
IL34	Down

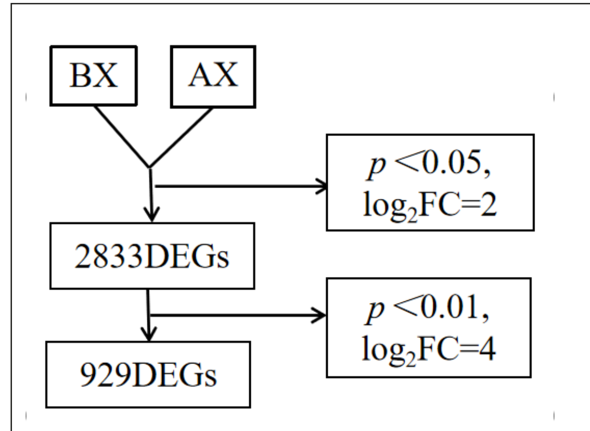

Figure 2. Flowchart for the identification of differentially expressed genes. A total of 2,833 genes exhibited variations with $p<0.05$ and $\log_2FC=2$. Upon differential screening, 929 genes exhibited variations with $p<0.01$ and $\log_2FC=4$.

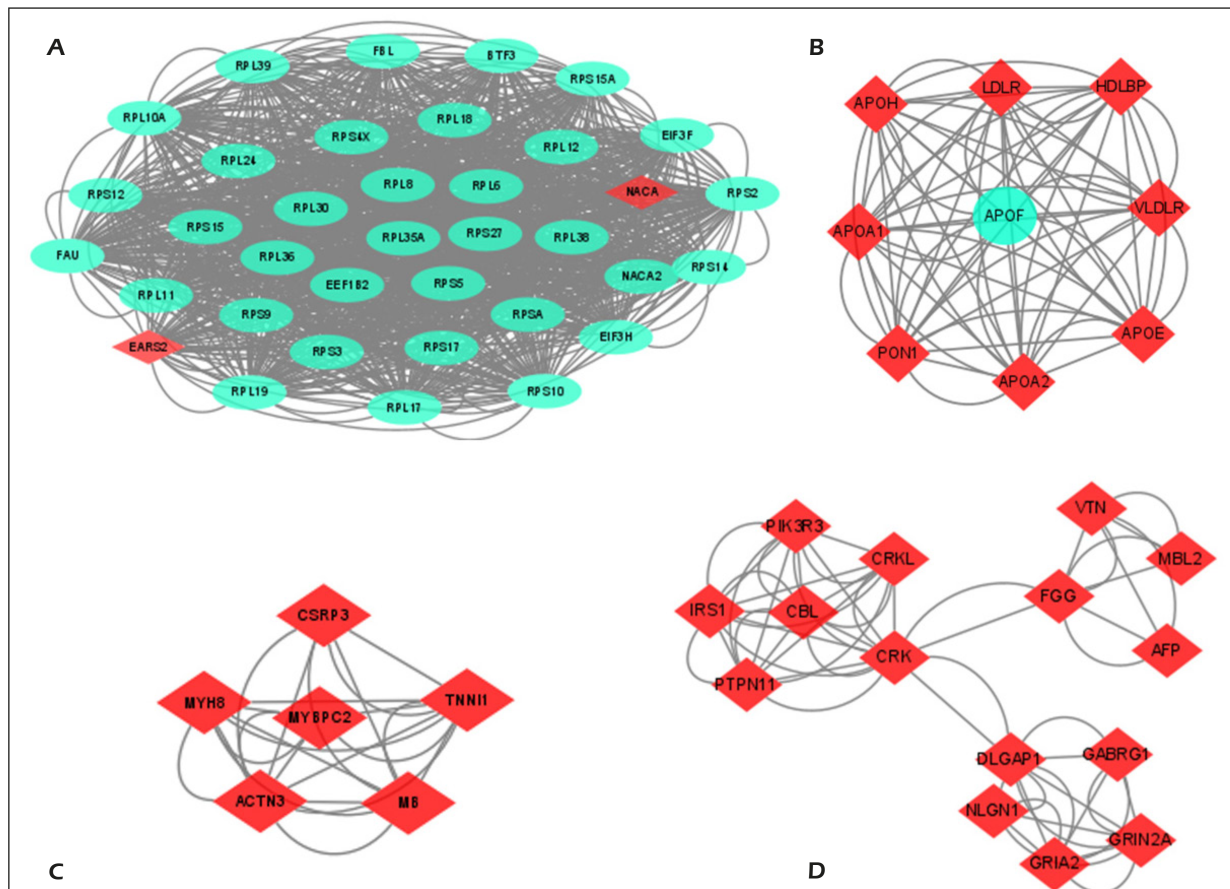

Figure 3. PPI network of DEGs and four cluster modules extracted by MCODE. **A,** The interaction network between proteins coded by DEGs. Each node represents a protein, while each edge represents one protein-protein association. Red diamonds represent the upregulated genes, and blue rectangle represent the downregulated genes (score: 34.914, 36 nodes and 1,222 edges). **B,** (score: 9, 9 nodes and 72 edges), followed by cluster 2. **C,** (score: 4.8, 6 nodes and 24 edges), cluster 3. **D,** (score: 4.429, 15 nodes and 42 edges), cluster 4.

browning, thereby reducing obesity[39,40]. Thus, the MAPK-signaling pathway occupies a crucial role in adipose mobilization. In our study, we found that exercise up-regulates *IGF-1* gene expression, and that up-regulation of *IGF-1* expression stimulates the *IGF-1* protein receptor, which activates key signaling proteins in the MAPK pathway[41], and the activation of the MAPK pathway is integral for *IGF-1* activity[42].

This studies also found that exercise can up-regulate the expression of FOXO-signaling pathway. Moreover, FOXO-signaling-pathway-related *G6PC* and *IGF-1* genes may reduce the efficiency of adipose-derived stem cell (ADSC) transplantation in T2DM, and FOXO family transcription factors also impede aspects of the aging process downstream of the insulin/IGF-signaling pathway. *IGF-1* has also been linked to the FOXO signaling pathway, increase in *IGF-I* can augment *FOXO1* content in muscle and adipose tissue.

Our GO analysis showed that exercise up-regulated lipid synthesis and metabolic pathways. Other studies[43] have revealed significantly enriched functions such as vasculature development, RNA processing, positive regulation of macromolecule metabolic process, blood vessel morphogenesis, negative regulation of gene expression, regulation of cell migration, and DNA replication extracellular matrix-receptor (ECM-receptor) interaction. The down-regulation process observed in this paper was determined to be an RNA catabolic process associated with ribosome and protein processes. Although ribosomal transcription and translation are closely related to adipocyte formation, the relationship between exercise and ribosomal down-regulation remains arcane.

Obesity is associated with inflammation[44], and inflammation is a key feature of type 2 diabetes[45]. This study compared the samples before and after exercise in three data sets, and found that the expression of *IL-1, IL-2, IL-3, IL-5, IL-17, IL-36* and other genes in adipose tissue was up-regulated, while the expression of *IL-34* was down-regulated. The up-regulated genes were mostly pro-inflammatory factors, and the down-regulated genes were mostly anti-inflammatory factors. It indicates that exercise changes the immune microenvironment of adipose tissue. The expression changes of pro-inflammatory and anti-inflammatory genes affect the inflammatory response of adipose tissue.

First, differential gene analysis of GSE43471 showed that the inflammatory factor *IL-17* was significantly up-regulated. Studies have shown that although *IL-17* is a pro-inflammatory factor, The *IL-17* pathway may also function in a negative-feedback fashion to inhibit the production of excess adipose tissue[46]. The inflammatory factor *IL-17* can inhibit adipose formation, regulate adipose tissue accumulation, and regulate glucose metabolism in mice[47], and *IL-17* antibody can reduce obesity by reducing the accumulation of macrophages in adipose tissue and reducing inflammation[48]. Down-regulated inflammatory factors include *IL-34*, which has been shown to enhance fat accumulation and inhibit the stimulating effect of insulin on glucose transport, which is related to insulin resistance[49]. The results based on this data set suggest that the body can reduce inflammation during moderate exercise. Secondly, analysis of the differential genes obtained from the samples in the GSE116801 dataset showed that there were no inflammatory factors. Therefore, this study speculated that due to the low exercise intensity, inflammation was not caused. Finally, the samples in the GSE58559 dataset were not analyzed owing to no pre-exercise sample. Comprehensive analysis of the three data sets showed that inflammatory factors significantly up-regulated were *IL-1*, *IL-2*, *IL-3*, *IL-5*, *IL-17, IL-36,* etc. When GSE43471 and GSE116801 were analyzed respectively, no above inflammatory factors were obtained. Therefore, it was speculated that *IL-1, IL-2, IL-3, IL-5* and *IL-36* were significantly up-regulated in the GSE58559 data set. It has been suggested that obesity can lead to pro-inflammatory states in metabolic tissues, such as up-regulation of pro-inflammatory cytokines TNFα and IL-1β, which can interfere with insulin signaling in adipocytes[50-53]. According to previous studies[54-56], *IL-1*, *IL-2*, *IL-3* and *IL-5* are mostly pro-inflammatory factors in the body, which suggests that the body is exercising vigorously, and the expression of pro-inflammatory factors is up-regulated, and inflammatory factors increase in adipose tissue. Strenuous exercise may promote the occurrence of inflammation in the body, and even lead to the imbalance of immune microenvironment in adipose tissue. IL-36 is a newly discovered cytokine and a new member of the IL-1 family, whose gene acts in a signaling pathway similar to that of traditional IL-1 family members[57]. Abnormal expression of *IL-36* may be related to the development of obesity.

Adipose tissue is also an immune organ, containing macrophages, granulocytes, lymphocytes and so on. The immune indexes of the body in-

clude immune cells (leukocytes, mononuclear phagocytes, T lymphocytes, etc.), immunoglobulin (IgA, IgG, IgM), cytokines (interferon, interleukin, tumor necrosis factor) and complement, etc. These immune indexes influence and restrict each other, and act on immune function together. Under normal circumstances, these immune cells maintain an anti-inflammatory balance in adipose tissue. These immune balances are disrupted when the body is obese. This brings to a chronic inflammatory condition of fat tissue, which leads to a decline in immune function. Immune cells such as macrophages and T cells infiltrating adipose tissue are the main sources of inflammatory factors such as TNF and IL-6[58], and skeletal muscle IL-6 may be involved in the anti-inflammatory effects of exercise-induced fat loss[59]. Macrophages have a bidirectional effect in the body, which can play an inflammatory response to phagocytose dead cells and viruses, etc., but their overreaction will trigger an over strong inflammatory response, causing harm to the body. During the formation of obesity, the position of macrophages in adipose tissue changes, and they change from anti-inflammatory phenotype to pro-inflammatory phenotype, that is, from M2 type to M1 type in obesity. At the same time, obesity leads to an overall decline in immune function. The proportion of CD8+ and CD4+ T cells increased in obese adipose tissue, while the number of immunosuppressed CD4+ regulatory T cells (known to secrete anti-inflammatory cytokines that inhibit macrophage migration) decreased. Macrophages accumulate in obese adipose tissue and secrete *TNF-α, IL-6, IL-1, McP-1* and *MIP-1α*[59-61].

The function of immune system is largely affected by physical exercise[62]. Regular physical activity can prevent and treat various chronic diseases associated with low-grade inflammation[63], leading to an increase in cytokines with anti-inflammatory properties at the systemic level[64]. At the same time, exercise may limit the fibrosis of white adipose tissue and the infiltration of pro-inflammatory immune cells[5].

Different types of exercise have different effects on immune cells. Studies[65] have shown that regular low-intensity and moderate-intensity exercise is related to the reduction of circulating pro-inflammatory markers and the improvement of immune function. Moderate-intensity aerobic exercise can promote the polarization of macrophages (M2-M1), thus relieving the inflammatory state of adipose tissue and reducing obesity[66]. At the same time, moderate intensity exercise can increase the number of natural killer (NK) cells, the number of neutrophils, eosinophils and lymphocytes in the circulating blood, and can increase the function of B cells. Recent studies[67] have shown that moderate and low intensity exercise can enhance neutrophil oxidation activity and macrophage phagocytosis, as well as increase the percentage of CD4+ T lymphocytes *IL-1β* content and reduce circulating TNF-α and IL-6 levels. Therefore, a long time of medium and low intensity exercise can play a significant role in immune defense. However, high-intensity exercise inhibits the function of many immune cells. Long-term high-intensity exercise can result in reduced T cell differentiation, reduced lymphocytes and monocytes, impaired neutrophil function, and reduced B cell function[68-70]. Meanwhile, prolonged and/or vigorous exercise has been found[71] to increase pro-inflammatory cytokines such as *IL-6, IL-8, TNF-α* and *IL-1*, and decrease NK cell, T and B lymphocyte and neutrophil activity.

Conclusions

In this study, bioinformatics was used to analyze the potential mechanism of exercise training-induced changes in obesity, and it was found that different exercise intensity can up-regulate MAPK signaling pathway and regulate *IGF-1* to induce adipose mobilization in adipose tissue, and the immune microenvironment in adipose tissue also changed with different exercise intensity. Although high-intensity exercise can strengthen the catabolism of fat in the body's adipose tissue, it also increases the occurrence of inflammation and leads to the imbalance of immune microenvironment again. Therefore, appropriate exercise intensity (moderate intensity and below) is the best way for the general population to reduce fat and weight.

Conflict of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interests.

Availability of Data and Materials

The (GSE datasets) data that support the findings of this study are available in the GEO database (https://www.ncbi.nlm.nih.gov/geo/).

Authors' Contributions

Conception and design of the work: Zhimin Lu and Zhiyuan Sun; acquisition and analysis of data: Sen Zhang and Xing Jiang; interpretation of data: Ling Ding and Chengzhi Li; writing and preparing the original draft: Zhimin Lu; writing, reviewing and editing the paper: Xuewen Tian and Qinglu Wang. All authors have read and agreed to the published version of the manuscript and to have agreed to both be personally accountable for the author's contributions and ensure to answer any questions related to the accuracy or integrity of any part of the work. All authors read and approved the final manuscript.

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