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CYTOKINE AND MATRIX METALLOPROTEINASE PROFILE IN THE SKELETAL MUSCLE OF RATS WITH DIET-INDUCED OBESITY

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Background. Our previous studies revealed impaired contractile performance of skeletal muscle in diet-induced obesity, accompanied by intramyocellular lipid accumulation and signs of fibrosis. These findings suggest that obesity-related muscle dysfunction may result from activation of pro-fibrotic pathways and imbalance in extracellular matrix turnover driven by chronic low-grade inflammation – a hallmark of obesity pathogenesis. This study aimed to evaluate the effects of long-term high-fat feeding on the cytokine profile and matrix remodeling markers in rat skeletal muscle and to identify possible biochemical mechanisms underlying obesity-related muscle dysfunction.

Materials and Methods. Adult male Wistar rats were fed either a standard chow (6.7 % fat; 15.27 kJ·g-1) or a high-fat diet (38.8 % fat; 28.71 kJ·g-1) for ten weeks. Serum biochemical parameters (total protein, glucose, cholesterol, bilirubin fractions, urea, creatinine, alanine and aspartate aminotransferases, α-amylase, alkaline phosphatase, γ-glutamyltransferase) were analyzed. Cytokines (TNF-α, IL-6, IL-1β, IL-10, IFN-γ), matrix metalloproteinases (MMP-2, MMP-9), and their inhibitor (TIMP-1) were quantified in skeletal muscle homogenates by ELISA.

Results. A high-fat diet produced pronounced alterations in serum biochemical parameters, reflecting profound metabolic dysregulation and systemic impairment of homeostatic control. In skeletal muscle, obesity induced a significant rise in pro-inflammatory cytokines (TNF-α, IL-6, IL-1β, IFN-γ) and a reduction in anti-inflammatory IL-10. These shifts were accompanied by increased MMP-2 and MMP-9 levels and a decline in TIMP-1, suggesting disturbed extracellular matrix turnover and early profibrotic remodeling.

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Conclusion. Diet-induced obesity promotes a sustained pro-inflammatory state accompanied by enhanced extracellular matrix remodeling. The imbalance between cytokines, metalloproteinases, and their inhibitor may underlie structural stiffening and decreased contractile efficiency of muscle fibers. A deeper understanding of these cytokine-MMP/TIMP interactions may provide new insights into the mechanisms of obesity-related muscle dysfunction and identify potential targets for maintaining skeletal muscle performance.

Keywords: obesity, skeletal muscle, cytokines, matrix metalloproteinases, extracellular matrix, inflammation, metabolic dysfunction

INTRODUCTION

Obesity is a chronic, multifactorial disease characterized by excessive adipose tissue accumulation and profound metabolic dysregulation (Wang *et al*., 2025). Beyond its well-established systemic consequences, obesity markedly affects skeletal muscle – a tissue that constitutes more than 30 % of total body mass and plays a central role in glucose homeostasis, fatty acid oxidation, and energy balance (Mengeste *et al*., 2021). Accumulating evidence indicates that skeletal muscle dysfunction in obesity extends beyond impaired contractility to involve biochemical, inflammatory, and structural remodeling processes that collectively contribute to metabolic inflexibility and reduced physical performance (Li *et al*., 2024; Castillo *et al*., 2025). However, despite extensive research, the precise molecular mechanisms linking metabolic alterations to impaired mechanical performance of skeletal muscle remain insufficiently defined.

Obesity is associated with a chronic, low-grade inflammatory state that disrupts tissue homeostasis across multiple organs (Cavaliere *et al*., 2023). Skeletal muscle, traditionally viewed as a purely mechanical system, is now recognized as an endocrine organ that secretes cytokines and myokines, influencing local and systemic metabolism (Guo *et al*., 2023). In obesity, excess lipid deposition within muscle fibers (intramyocellular lipids) triggers local activation of pro-inflammatory signaling pathways, including NF-κB and JNK cascades, resulting in elevated expression of pro-inflammatory cytokines and various chemokines that perpetuate local inflammation and immune cell recruitment in skeletal muscle (Jia & Sowers, 2019). This persistent inflammatory signaling promotes mitochondrial stress and impairs tissue regeneration, leading to progressive myocyte damage and fibrosis (Li *et al*., 2024; Catalán *et al*., 2018). At the same time, data from different studies remain partially inconsistent regarding the extent and functional consequences of cytokine alterations in skeletal muscle, suggesting the need for more integrated analyses combining inflammatory and structural markers.

Oxidative stress represents another major pathogenic mechanism in obesity-related muscle dysfunction. Long-term high-calorie intake and lipid excess enhance reactive oxygen species (ROS) generation, leading to oxidation of proteins and lipids and dysregulation of calcium handling. Experimental studies have demonstrated that high-fat feeding induces excessive production of ROS in skeletal muscle, accompanied by marked mitochondrial morphological alterations, such as swelling and fragmentation, which are closely associated with the development of insulin resistance and apoptotic signaling (Lee *et al*., 2021). Pro-inflammatory cytokines such as TNF-α and IL-6 further exacerbate these effects by downregulating mitochondrial fusion proteins like mitofusin-2, thereby impairing oxidative phosphorylation efficiency and ATP generation (Bach *et al*., 2005).

Beyond these metabolic and inflammatory disturbances, obesity also induces pronounced structural changes in skeletal muscle, which in turn compromise its contractile function and mechanical performance (Bollinger, 2017). Among the key factors driving these morphological changes is the excessive accumulation of intramuscular lipid inclusions (Jia & Sowers, 2019). The buildup of such lipid depots represents a hallmark morphological manifestation of obesity-related muscle pathology. Lipid inclusions possess greater stiffness compared with contractile fibers, thereby altering the mechanical properties of the muscle and reducing both its elasticity and efficiency of force transmission during contraction (Rahemi *et al*., 2015).

Structural remodeling in obese skeletal muscle involves not only lipid infiltration but also the development of interstitial fibrosis (Li *et al*., 2024). Matrix metalloproteinases (MMPs) together with their endogenous tissue inhibitors (TIMPs) serve as principal regulators of collagen degradation and extracellular matrix remodeling. Persistent low-grade inflammation disrupts the MMP/TIMP equilibrium, leading to excessive collagen accumulation in intermuscular connective tissue, restricted sarcomere mobility, and accelerated development of muscle stiffness (Alameddine & Morgan, 2016). Despite the recognized role of MMP/TIMP imbalance in tissue remodeling, their contribution to skeletal muscle dysfunction in obesity, particularly in relation to inflammatory signaling, remains insufficiently explored.

Taken together, these findings indicate that obesity profoundly alters the metabolic, inflammatory, and structural integrity of skeletal muscle, leading to impaired contractile performance. In our previous studies, we demonstrated that diet-induced obesity is associated with significant alterations in mechanokinetic parameters of skeletal muscle contraction, including changes in force generation and fatigue development, indicating impaired functional capacity of the *musculus soleus* (Nozdrenko *et al*., 2026). However, the molecular mechanisms underlying these functional disturbances remain unclear. Building upon these findings, the present study aims to analyze biochemical and molecular markers to identify possible links between inflammation, extracellular matrix (ECM) remodeling, and muscle dysfunction. By integrating cytokine profiling with the assessment of MMP/TIMP system components, this work seeks to provide a more comprehensive understanding of the mechanisms connecting inflammation, structural remodeling, and impaired muscle performance in obesity.

MATERIALS AND METHODS

Animals and experimental design. Experiments were performed on forty adult male Wistar rats with an initial body weight of 135 ± 10 g. All animal procedures were approved by the Ethical Committee of Taras Shevchenko National University of Kyiv (Protocol No. 7, dated 21.11.2024) and conducted in accordance with the European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes (Council of Europe, 1997), as well as national legislation and institutional ethical standards.

Prior to the experiment, animals were allowed to acclimate for seven days under controlled environmental conditions: temperature 22 ± 3 °C, relative humidity 60 ± 5 %, and a 12 h light/12 h dark cycle. Standard rodent chow and water were provided *ad libitum* during the acclimation period.

After acclimatization, animals were divided equally into two groups (n = 20): Control and Obesity. Rats were housed in polypropylene cages (five animals per cage) under the same vivarium conditions, with free access to water and their assigned diet for ten weeks.

Rats in the Control group were fed a standard laboratory chow diet (6.7 % fat, 21 % protein, and 55.1 % carbohydrates; energy content 15.27 kJ·g^{-1}; LabDiet 5001, USA). Rats in the Obesity group were fed a high-fat diet (HFD) containing 38.8 % fat, 15.5% protein, and 45.7 % carbohydrates (energy content 28.71 kJ·g^{-1}). The HFD was prepared in-house by supplementing standard chow (60%) with pork fat (10 %), eggs (10 %), sucrose (9 %), dry milk (5 %), peanuts (5 %), and vegetable oil (1 %) (Shen *et al*., 2010). The diet-induced obesity model used in this study has been previously characterized in our earlier investigations, which demonstrated significant metabolic disturbances (Halenova *et al*., 2018), lipid accumulation in skeletal muscle, and structural alterations of muscle tissue under conditions of prolonged high-fat feeding (Nozdrenko *et al*., 2026).

Sample preparation. At the end of the 10th week, the animals were fasted overnight and humanely euthanized by cervical dislocation. Blood was collected by cardiac puncture under sterile conditions and allowed to clot for 30 min at 37 °C. Samples were centrifuged at 2500 g for 15 min, and the serum was collected into sterile microtubes. The obtained serum was stored at −20 °C until biochemical assays were performed.

To determine local cytokine and matrix metalloproteinase levels, skeletal muscle samples were collected immediately after euthanasia. Tissue was rinsed in ice-cold physiological saline, cleared of connective tissue, weighed, and homogenized in a 1:10 (w/v) ratio in 0.05 M Tris-HCl buffer (pH 7.4) containing 0.13 M NaCl. Homogenization was carried out using an automatic homogenizer FSH-2A (Vevor, Poland) at 13,000 rpm for 2 min under cooled conditions. The homogenate was centrifuged at 10,000 g for 15 min at +4 °C, and the supernatant was collected into sterile microtubes and stored at −20 °C for further biochemical analysis.

Biochemical analysis of some blood metabolic parameters. Serum biochemical parameters were measured using a semi-automatic biochemical analyzer Humalyzer 3000 (Human GmbH, Germany). The following indicators were measured: total protein, glucose, total cholesterol, total, direct and indirect bilirubin, urea, creatinine, alanine aminotransferase (ALT), aspartate aminotransferase (AST), α-amylase, alkaline phosphatase, and γ-glutamyltransferase (GGT). The concentration of glycosylated hemoglobin in the blood of experimental rats was measured using standard kits manufactured by PLIVA-Lachema Diagnostika (Czech Republic). Serum insulin levels were determined using a commercial ELISA kit (XEMA, Ukraine) according to the manufacturer's instructions.

Enzyme-linked immunosorbent assay (ELISA). The levels of cytokines (interleukin (IL)-6, -1β, -10, tumor necrosis factor-α (TNF-α), and interferon-γ (IFN-γ)) as well as matrix metalloproteinases (MMP-2 and MMP-9), and their tissue inhibitor-1 (TIMP-1) in skeletal muscle homogenates were measured using ELISA following standard protocols. Prior to analysis, total protein concentration in each homogenate was measured, and samples were adjusted to an equal protein concentration (10 μg·mL^{-1}) with 0.05 M Tris-HCl buffer (pH 7.4) containing 0.13 M NaCl and added to 96-well microplates (Thermo Fisher Scientific, USA) at 100 μL per well, followed by overnight incubation at 4 °C to allow antigen adsorption. This approach ensured equal loading of total protein across all wells, allowing reliable comparison of target protein levels between experimental groups. Plates were then washed three times with wash buffer containing 0.05 % Tween-20 and blocked with 3 % nonfat dry milk for 1 h at 37 °C. After washing, primary

antibodies (Santa Cruz Biotechnology, USA) were added and incubated for 1 h at 37 °C, followed by horseradish peroxidase (HRP)-conjugated secondary antibodies (Sigma-Aldrich, USA) for 1 h under the same conditions. After washing, o-phenylenediamine (OPD) substrate with hydrogen peroxide was added, and the enzymatic reaction was stopped with 2.5 N H_2SO_4. The optical density was measured at 492 nm using a μQuantTM microplate reader (BioTek Instruments, USA). The results were expressed as relative units per mg of protein.

Total protein concentration was determined using the Quick Start™ Bradford Protein Assay (Bio-Rad Laboratories, USA) according to the manufacturer's instructions.

Statistical analysis. Statistical analysis was performed using GraphPad Prism 8.0 software (GraphPad Software, USA). The normality of data distribution was assessed using the Shapiro–Wilk test. For parameters showing normal distribution, intergroup comparisons between the Control and Obesity groups (n = 20 per group) were performed using a two-tailed unpaired Student's t-test with Welch's correction for unequal variances. For variables that did not meet the assumption of normality, the nonparametric Mann-Whitney U test was applied. All results are presented as mean ± standard deviation (M ± SD). Differences were considered statistically significant at $p < 0.05$.

RESULTS AND DISCUSSION

Excessive caloric intake is one of the primary factors driving systemic metabolic dysregulation in obesity. Prolonged overnutrition leads to lipid accumulation not only in adipose tissue but also in non-adipose organs such as skeletal muscle and liver, contributing to lipotoxicity and metabolic imbalance (Cavaliere *et al*., 2023; Wang *et al*., 2025). Alterations in serum biochemical parameters provide an integrated view of these systemic changes, reflecting perturbations in carbohydrate, lipid, and protein metabolism as well as hepatic and renal function (Borovkov *et al*., 2024).

As shown in **Table**, a high-fat diet induced pronounced alterations in the biochemical profile of the obese rats compared with controls. A significant rise in serum glucose and total cholesterol levels may indicate disturbances in carbohydrate and lipid metabolism, which are well-recognized features of obesity-related metabolic imbalance (Borovkov *et al*., 2024; Mika & Sledzinski, 2017). In addition, elevated levels of glycosylated hemoglobin and insulin were observed in obese animals, reflecting sustained hyperglycemia and altered insulin signaling. Together, these changes may suggest the development of insulin resistance under prolonged high-fat feeding. The elevation in total protein concentration may indicate enhanced hepatic synthesis or early adaptive responses to metabolic stress.

Changes in bilirubin fractions (see **Table**) may reflect alterations in hepatic heme metabolism rather than direct evidence of hepatocellular dysfunction, consistent with metabolic adaptations accompanying diet-induced obesity (Soltis *et al*., 2017). On the other hand, considering the antioxidant and signaling functions of bilirubin, its decrease may represent an adaptive hepatic response to oxidative stress associated with obesity (Žiberna *et al*., 2021).

Renal function markers demonstrated a modest but significant increase in serum creatinine, whereas urea levels remained unchanged, suggesting early functional stress on glomerular filtration rather than overt renal injury.

Among enzymatic indicators of metabolic activity, α-amylase and alkaline phosphatase were markedly elevated in obese animals (see **Table**), reflecting increased pancreatic and hepatobiliary activity associated with high lipid and carbohydrate turnover. Although alanine aminotransferase (ALT) activity showed no significant difference, a reduction in aspartate aminotransferase (AST) may be associated with altered metabolic activity or mitochondrial function under conditions of lipid accumulation in hepatocytes. Similar findings were reported by S. B. Borovkov *et al*. (2024), who observed reduced AST activity associated with disrupted bile secretion and hepatic metabolic alterations in obese animals. A notable decrease in γ-glutamyltransferase (GGT) activity may also reflect changes in hepatic metabolic regulation rather than impaired liver function per se.

Serum biochemical parameters in control and obese rats

Parameter	Control (*n* = 20)	Obesity (*n* = 20)	*p*-value
Total protein, g·L^{-1}	76.20 ± 4.44	88.80 ± 2.59	0.0012
Glucose, mmol·L^{-1}	4.30 ± 0.32	6.46 ± 0.36	<0.0001
Glycosylated hemoglobin, μmol fructosyl residues·g^{-1} hemoglobin	0.21±0.03	0.81±0.06	0.012
Insulin, rel. units·mg^{-1} protein	0.13±0.01	0.26±0.02	0.024
Total cholesterol, mmol·L^{-1}	1.96 ± 0.23	3.48 ± 0.34	<0.0001
Total bilirubin, μmol·L^{-1}	10.34 ± 0.34	8.84 ± 0.68	0.005
Direct bilirubin, μmol·L^{-1}	1.48 ± 0.18	1.44 ± 0.05	0.6538
Indirect bilirubin, μmol·L^{-1}	9.06 ± 0.54	7.40 ± 0.67	0.003
Urea, mmol·L^{-1}	6.32 ± 0.30	5.88 ± 0.70	0.248
Creatinine, μmol·L^{-1}	50.60 ± 1.95	59.20 ± 1.92	0.0001
α-Amylase, U·L^{-1}	636.00 ± 32.63	758.40 ± 40.87	0.007
Alkaline phosphatase, U·L^{-1}	126.80 ± 50.47	407.80 ± 33.12	<0.0001
ALT, nmol·L^{-1}	79.60 ± 14.48	69.20 ± 5.81	0.194
AST, nmol·L^{-1}	260.80 ± 10.96	218.80 ± 28.76	0.0274
GGT, U·L^{-1}	23.00 ± 3.16	11.80 ± 1.48	0.0005

Note: ALT – alanine aminotransferase, AST – aspartate aminotransferase, GGT – γ-glutamyltransferase; data are presented as mean ± SD; values of $p < 0.05$ were considered statistically significant

Overall, these data confirm that sustained high-fat feeding disrupts systemic metabolic homeostasis, producing coordinated alterations in glucose/lipid metabolism as well as enzyme activity that collectively mirror the early stages of obesity-associated metabolic dysfunction.

Long-term exposure to a high-fat diet induces a chronic low-grade inflammation within skeletal muscle, driven by cytokines produced by infiltrating macrophages and resident myocytes (Cavaliere *et al*., 2023). The obese group exhibited a significant elevation in the levels of TNF-α (**Fig. 1*A***), IL-6 (**Fig. 1*B***), IL-1β (**Fig. 1*C***), and IFN-γ (**Fig. 1*D***), accompanied by a marked reduction in the anti-inflammatory cytokine IL-10 (**Fig. 1*E***) compared with control animals. This pattern reflects a shift toward a persistent pro-inflammatory state within muscle tissue, a hallmark of obesity-related metabolic dysfunction.

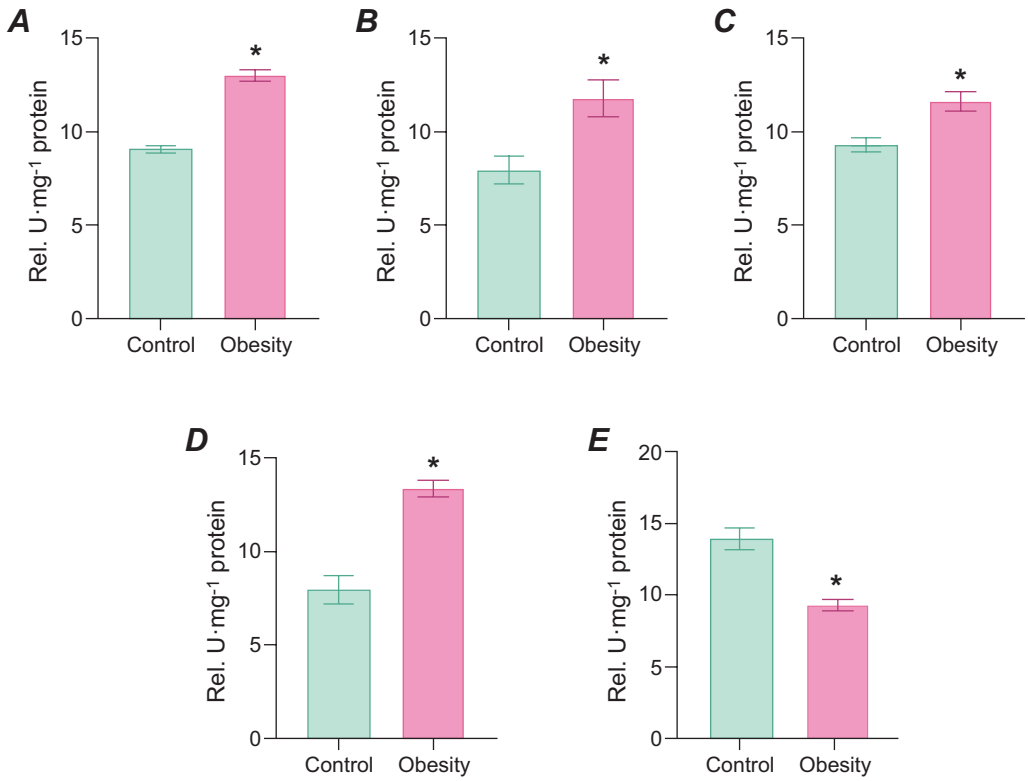

Fig. 1. Levels of TNF-α (***A***) IL-6 (***B***), IL-1β (***C***), IFN-γ (***D***), and IL-10 (***E***) in skeletal muscle homogenates of control and obese rats. Data are presented as M ± SD (n = 20 per group); *$p < 0.05$ compared with the control group

Under physiological conditions, IL-6 functions as a myokine supporting glucose uptake and fatty acid oxidation, thereby maintaining skeletal muscle metabolic flexibility (Lin *et al*., 2023). However, under chronic nutrient excess, excessive IL-6 production promotes catabolic and fibrotic signaling, contributing to muscle metabolic inflexibility and structural remodeling (Muñoz-Cánoves *et al*., 2013; Kistner *et al*., 2022). The synergistic action of TNF-α and IL-1β further amplifies IL-6 transcription, sustaining a pro-inflammatory microenvironment within myofibers.

The marked increase in TNF-α, IL-6, and IL-1β observed in obesity (**Fig. 1*A*, *B*,** and ***C***, respectively) underscores their central role in mediating both metabolic and structural alterations in skeletal muscle. These cytokines are potent activators of NF-κB and JNK signaling, leading to impaired insulin signaling, mitochondrial oxidative stress, and increased expression of MMPs, thereby promoting extracellular matrix (ECM) remodeling (Li *et al*., 2024; Webster *et al*., 2020). Sustained cytokine activity also stimulates fibroblast proliferation and collagen synthesis via TGF-β and integrin-mediated pathways, contributing to ECM deposition and tissue stiffness (Musale *et al*., 2023).

In addition to TNF-α, IL-6, and IL-1β, the elevated levels of IFN-γ (**Fig. 1*D***) observed in obese skeletal muscle confirm the activation of Th1-type immune responses. Acting together with TNF-α and IL-1β, IFN-γ enhances M1 macrophage polarization and chemokine expression, thereby promoting immune cell recruitment and sustaining chronic inflammation (Webster *et al*., 2020; Babaeijandaghi *et al*., 2022).

Collectively, this cytokine network creates a self-perpetuating inflammatory loop in which metabolic, inflammatory, and fibrotic pathways reinforce one another, linking chronic inflammation to impaired muscle regeneration and function.

In contrast, the reduction in IL-10 (**Fig. 1*E***) observed in obese rats indicates a weakening of endogenous anti-inflammatory control. IL-10 normally suppresses TNF-α and IL-1β synthesis and limits NF-κB activation; however, chronic metabolic overload likely diminishes IL-10 expression and receptor signaling, lowering its protective efficacy (Toita *et al*., 2016). The resulting dominance of pro-inflammatory mediators promotes oxidative stress, myofiber injury, and defective tissue regeneration.

Given the established role of inflammatory cytokines in regulating ECM turnover, particular attention was directed to matrix metalloproteinases (MMPs) as downstream mediators of cytokine-driven remodeling.

As shown in **Fig. 2**, the obese group exhibited significantly increased levels of MMP-2 and MMP-9 in skeletal muscle homogenates, along with a pronounced decrease in their inhibitor TIMP-1, compared with controls. This pattern suggests an imbalance between ECM degradation and synthesis rather than a simple activation of proteolysis.

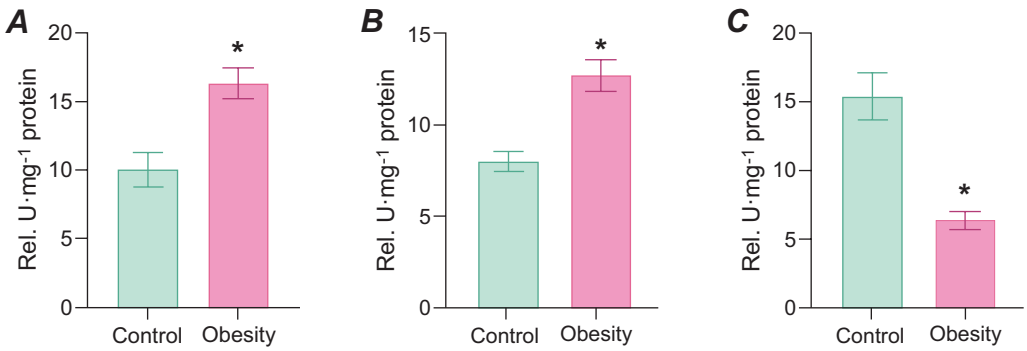

Fig. 2. Levels of MMP-2 (***A***), MMP-9 (***B***), and TIMP-1 (***C***) in skeletal muscle homogenates of control and obese rats. Data are presented as M ± SD (n = 20 per group); * – $p < 0.05$ compared with the control group

Elevated MMP-2 and MMP-9 levels are closely linked to systemic inflammation and metabolic dysregulation in obesity. These gelatinases degrade collagens, elastin,

and fibronectin, compromising structural integrity and facilitating immune and fibroblast infiltration into the interstitium (Wiśniewski *et al*., 2024). The resulting macrophage accumulation amplifies the release of TNF-α, IL-6, and IL-1β, further upregulating MMP transcription and creating a chronic inflammatory cycle (Alameddine & Morgan, 2016; Musale *et al*., 2023).

Beyond their structural role, MMP-2 and MMP-9 are implicated in insulin resistance. By promoting ECM remodeling and inflammatory cell infiltration, they contribute to local hypoxia, oxidative stress, and activation of JNK and p38 MAPK, which suppress insulin receptor substrate phosphorylation and downstream Akt signaling (Ruiz-Ojeda *et al*., 2019; Wiśniewski *et al*., 2024). Persistent activation of these stress pathways leads to reduced glucose uptake and mitochondrial dysfunction – biochemical hallmarks of muscle insulin resistance.

Furthermore, excessive ECM degradation releases bioactive fragments (matrikines) that act as paracrine mediators, stimulating fibroblast activation and collagen deposition. This process establishes a profibrotic environment, increasing tissue stiffness and compromising muscle contractile properties (Alameddine & Morgan, 2016; Musale *et al*., 2023).

Collectively, these findings suggest that sustained MMP-2 and MMP-9 upregulation not only disturbs ECM homeostasis but also integrates inflammatory, metabolic, and mechanical alterations underlying skeletal muscle dysfunction in obesity.

The reduction of TIMP-1 observed in obese animals (**Fig. 2*C***) further exacerbates this proteolytic-fibrotic imbalance. Under physiological conditions, TIMP-1 tightly regulates MMP activity to preserve ECM integrity. However, prolonged exposure to TNF-α and IFN-γ may suppress TIMP-1 transcription, thereby reducing its inhibitory capacity and permitting uncontrolled matrix turnover (Musale *et al*., 2023).

The biochemical alterations observed in the present study are in line with structural changes previously reported under identical experimental conditions. In our earlier work using the same diet-induced obesity model, histological analysis of the *musculus soleus* revealed the accumulation of intramyocellular lipid droplets and increased deposition of intramuscular collagen fibers, indicating early fibrotic remodeling of skeletal muscle tissue (Nozdrenko *et al*., 2026). These structural changes were accompanied by impaired contractile performance and elevated biochemical markers of muscle damage. Together, these findings support the concept that chronic inflammation and extracellular matrix remodeling are key mechanisms contributing to obesity-associated skeletal muscle dysfunction.

CONCLUSION

Taken together, these findings indicate that diet-induced obesity profoundly alters the cytokine and proteolytic regulatory networks in skeletal muscle, shifting them toward a persistent pro-inflammatory and pro-fibrotic state. The observed upregulation of TNF-α, IL-6, IL-1β, IFN-γ, MMP-2, and MMP-9, together with a decline in IL-10 and TIMP-1, reflects an imbalance between inflammatory and reparative signaling that disturbs extracellular matrix homeostasis and impairs metabolic flexibility.

The obtained results provide an important framework for understanding how chronic cytokine activation and matrix remodeling converge with oxidative stress and fibroblast activation, ultimately altering myofiber structure and reducing contractile

performance under obesity conditions. Integration of these biochemical data with our previous mechanokinetic analysis of the *musculus soleus* (Nozdrenko *et al*., 2026) suggests that the loss of muscle elasticity and the accelerated development of fatigue in obesity are closely linked to cytokine-driven ECM reorganization and metabolic rigidity.

A deeper understanding of the cytokine-MMP/TIMP regulatory network may help clarify how inflammatory and structural alterations interact to disrupt muscle contractile dynamics and contribute to obesity-related decline in mechanical performance. Moreover, elucidating these molecular interactions may reveal potential targets for controlling pathological ECM remodeling and preserving skeletal muscle functionality in metabolic disorders.

COMPLIANCE WITH ETHICAL STANDARDS

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 interest.

Animal studies: all institutional, national, and international guidelines for the care and use of laboratory animals were followed.

Human Rights: this article does not contain any studies with human subjects performed by any of the authors.

AUTHOR CONTRIBUTIONS

Conceptualization, [T.H.; Y.P.]; methodology, [T.H.; N.R.]; investigation, [O.R., M.K., O.L.]; resources, [O.R., M.K.]; formal analysis, [T.H.; O.L.; N.R.]; writing – original draft preparation, [T.H.; O.R.]; writing – review and editing, [O.L.; N.R.; Y.P.]; visualization, [T.H.]; supervision, [Y.P.].

All authors have read and agreed to the published version of the manuscript.

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ПРОФІЛЬ ЦИТОКІНІВ І МАТРИКСНИХ МЕТАЛОПРОТЕЇНАЗ У СКЕЛЕТНИХ М'ЯЗАХ ЩУРІВ ЗА УМОВ ОЖИРІННЯ, ІНДУКОВАНОГО ДІЄТОЮ З ВИСОКИМ ВМІСТОМ ЖИРУ

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Вступ. У наших попередніх дослідженнях показано порушення скоротливої активності скелетних м'язів за умов ожиріння, індукованого тривалим споживанням дієти з високим вмістом жиру, що супроводжувалося накопиченням внутрішньоклітинних ліпідних включень та ознаками фіброзу. Отримані результати дають підстави припускати, що розвиток м'язової дисфункції за ожиріння пов'язаний з активацією профібротичних сигнальних каскадів і порушенням метаболізму позаклітинного матриксу, що формується під впливом хронічного запалення – характерної патогенетичної ознаки ожиріння. Метою роботи було оцінити вплив тривалого споживання високожирової дієти на профіль цитокінів і на маркери ремоделювання матриксу у скелетних м'язах щурів, а також визначити ймовірні біохімічні механізми розвитку м'язової дисфункції, асоційованої з ожирінням.

Матеріали та методи. Дорослих самців щурів лінії Вістар утримували протягом десяти тижнів на стандартному раціоні (6,7 % жиру; 15,27 кДж·г$^{-1}$) або на дієті з високим вмістом жиру (38,8 % жиру; 28,71 кДж·г$^{-1}$). У сироватці крові визначали біохімічні показники (загальний білок, глюкозу, холестерин, фракції білірубіну, сечовину, креатинін, активність аланін- і аспартатамінотрансфераз, α-амілази, лужної фосфатази та γ-глутамілтрансферази). Рівні цитокінів (TNF-α, IL-6, IL-1β, IL-10, IFN-γ), матриксних металопротеїназ (MMP-2, MMP-9) та їхнього інгібітора (TIMP-1) у гомогенатах м'язової тканини визначали методом імуноферментного аналізу.

Результати. Високожирова дієта спричиняла виражені зміни біохімічних показників сироватки крові, що відображало глибоке порушення метаболічної регуляції та гомеостазу на системному рівні. У скелетних м'язах ожиріння супроводжувалося зростанням рівнів прозапальних цитокінів (TNF-α, IL-6, IL-1β, IFN-γ) і зниженням антизапального IL-10. Встановлено підвищення рівнів MMP-2 і MMP-9 за одночасного зменшення TIMP-1, що може свідчити про дисбаланс обміну компонентів позаклітинного матриксу та початкові прояви профібротичного ремоделювання.

Висновки. Ожиріння, індуковане тривалим споживанням дієти з високим вмістом жиру, формує стійкий прозапальний стан у скелетних м'язах, що супроводжується посиленням процесів ремоделювання позаклітинного матриксу. Дисбаланс

між цитокінами, металопротеїназами та їхнім інгібітором може лежати в основі структурного ущільнення тканини і зниження скоротливої ефективності м'язових волокон. Глибше розуміння взаємодії цитокінів і MMP/TIMP може сприяти з'ясуванню механізмів розвитку м'язової дисфункції за ожиріння та визначенню потенційних мішеней для збереження функціональної активності скелетних м'язів.

Ключові слова: ожиріння, скелетна м'язова тканина, цитокіни, матриксні металопротеїнази, позаклітинний матрикс, запалення, метаболічна дисфункція

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