World Bulletin of Public Health (WBPH)
Available Online at: https://www.scholarexpress.net
Volume-41, December 2024
ISSN: 2749-3644

MATRIX METALLOPROTEINASES AND THEIR INHIBITORS IN PATIENTS WITH SCLERODERMA.

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Article history:

Received: September 28th 2024
Accepted: October 26th 2024

Abstract:

Scleroderma (SD) holds a significant place in modern dermatology, ranking second in prevalence among dermatological conditions. However, its exact pathogenesis remains insufficiently understood. One proposed primary mechanism for the development of sclerotic changes in the skin involves the activation of fibrogenesis, which is thought to stem from dysfunction within the extracellular matrix (ECM). The ECM is composed of structural proteins such as collagen, elastin, proteoglycans, and glycoproteins, all of which are regulated by a specific class of proteolytic enzymes called matrix metalloproteinases (MMPs). These enzymes are critical for maintaining ECM homeostasis and influence key biological molecules, including cytokines, interleukin-1β (IL-1β), tumor necrosis factor-α (TNF-α), transforming growth factor β1 (TGF-β1), and vascular endothelial growth factor (VEGF).

Keywords: Scleroderma

Scleroderma (SD) holds a significant place in modern dermatology, ranking second in prevalence among dermatological conditions. However, its exact pathogenesis remains insufficiently understood. One proposed primary mechanism for the development of sclerotic changes in the skin involves the activation of fibrogenesis, which is thought to stem from dysfunction within the extracellular matrix (ECM). The ECM is composed of structural proteins such as collagen, elastin, proteoglycans, and glycoproteins, all of which are regulated by a specific class of proteolytic enzymes called matrix metalloproteinases (MMPs). These enzymes are critical for maintaining ECM homeostasis and influence key biological molecules, including cytokines, interleukin-1β (IL-1β), tumor necrosis factor-α (TNF-α), transforming growth factor β1 (TGF-β1), and vascular endothelial growth factor (VEGF). These molecules mediate essential metabolic processes such as cellular proliferation, differentiation, migration, apoptosis, and angiogenesis. Additionally, MMPs play a crucial role in preserving the elasticity and structural integrity of vascular walls by breaking down components like denatured collagen, fibronectin, laminin, elastin, and vitronectin. This degradation facilitates component recycling and the formation of new, functional molecules. Specific types of MMPs have distinct roles. Gelatinases, including MMP-2 and MMP-9, hydrolyze type IV fibrillar collagen, while MMP-7 (elastase) is secreted by vascular and inflammatory cells such as macrophages and polymorphonuclear neutrophils. Elevated activity levels of MMP-2, MMP-9, and MMP-7 are closely associated with angiogenesis and apoptosis. Under normal physiological conditions, MMP expression is tightly regulated by tissue inhibitors of metalloproteinases (TIMPs), which differ in their selectivity for various metalloproteinases. For instance, TIMP-1 predominantly inhibits MMP-9 while TIMP-2 primarily targets MMP-2. Similarly, TIMP-3 and TGF-β1 act together to suppress MMP-7 activity. Furthermore, the inhibitory effects of TGF-β1 on MMP catabolism are reinforced by connective tissue growth factor, which enhances TIMP-1 and TIMP-3 expression while downregulating MMP-2 activity. Due to their relatively small molecular size and solubility in biological fluids, MMPs and TIMPs circulate in the bloodstream in concentrations reflective of tissue-level expression. This characteristic makes them readily detectable in blood serum. Over recent years, various MMPs, TIMPs, and TGF-β1 have gained attention as potential diagnostic and prognostic biomarkers. Shifts in the activity of these markers toward normal levels can indicate clinical recovery. Despite this progress, the systemic activity of MMPs and TIMPs in patients with scleroderma remains inadequately explored. Investigating the interplay between ECM factors and TGF-β1 may significantly enhance our understanding of scleroderma pathogenesis and provide a foundation for developing targeted therapeutic approaches. This study aimed to perform a comparative analysis of the levels of MMP-2, MMP-9, and MMP-7 alongside their inhibitors TIMP-1 and TIMP-2 in the blood serum of patients with systemic and localized forms of scleroderma.

MATERIAL AND METHODS OF A RESEARCH

A total of 64 patients participated in the study, comprising 29 women diagnosed with systemic scleroderma (SSD) and 35 individuals with localized scleroderma (LSD), including 26 women and 9 men. Additionally, 20 healthy donors were included as a

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control group, matched by age. The patients had a mean age of 55.3 ± 6.2 years, with an average disease duration of 13.5 ± 4.2 years. The study involved a thorough review of medical histories and a detailed physical examination using standard clinical and laboratory methods. When required, supplementary diagnostic procedures such as ultrasound imaging, rheovasography, endoscopy, echocardiography, and duplex ultrasonography were implemented. SSD diagnoses were confirmed based on the diagnostic criteria established by N.G. Guseva. Among the SSD patients, 20 (69%) exhibited chronic disease progression, while 9 (31%) demonstrated a subacute course. Moderate inflammatory activity was observed in 19 (65.5%) patients, whereas 10 (34.5%) showed minimal activity. Clinically, SSD presented with multisystem involvement affecting diverse organs, tissues, and systems. In contrast, all LSD cases were characterized by plaque formation. Of these, 16 patients (45.7%) were in the consolidation stage, while 19 (54.3%) were in the atrophic stage. Clinical, laboratory, and instrumental assessments in LSD patients revealed no signs of systemic involvement. Extracellular matrix enzyme levels and TGF-β1 concentrations were measured using a solid-phase ELISA technique on an AT-858 microplate reader (LTD, China). MMP and TIMP levels were quantified with standardized ELISA kits: "Human/Mouse/Rat MMP-2 (total)," "Human/Mouse/Rat MMP-9 (total)," "Human MMP-7 (total)," "Human TIMP-1," and "Human TIMP-2" (Quantikine, R&D Systems, USA). TGF-β1 levels were assessed using a commercial test kit from Bender MedSystems, Austria. All assays adhered strictly to the manufacturers' protocols. The concentrations of the studied parameters in serum samples were reported in nanograms per milliliter (ng/mL). Statistical analysis was conducted using the Statistica V.7 software. Mean values (M) and their standard errors (m) were calculated. Differences in data were evaluated using Student's t-test, and Pearson's correlation coefficient (r) was applied for correlation analysis. Statistical significance was established at $p < 0.05$

RESULTS AND DISCUSSION

The analysis of the results indicated that in patients with systemic sclerosis (SS), the activity of all measured matrix metalloproteinases (MMPs)—MMP-2, MMP-7, and MMP-9—was considerably lower, whereas the levels of tissue inhibitors of metalloproteinases (TIMPs), specifically TIMP-1 and TIMP-2, were higher compared to the control group (refer to the table). Furthermore, in patients with systemic scleroderma (SSD), the concentrations of MMP-2, MMP-7, and MMP-9 decreased by 17.1%, 13.9%, and 18.3% ($P<0.05$), respectively, while TIMP-1 and TIMP-2 levels rose by 18.2% and 19.5% ($P<0.05$) in comparison to individuals with localized scleroderma (LSD). Considering the pivotal role of MMPs and TIMPs in the regulation of fibrogenesis, serum levels of transforming growth factor beta 1 (TGF-β1), a key mediator of the process, were also assessed. TGF-β1 is known to drive myofibroblast differentiation, promote a fibrogenic phenotype in fibroblasts, regulate the production of platelet-derived growth factor (PDGF), and stimulate the synthesis of extracellular matrix components such as collagen and fibronectin. The findings revealed that SS patients exhibited significantly higher systemic concentrations of TGF-β1 compared to the control group. Moreover, TGF-β1 levels in SS patients exceeded those in SSD patients by 98.3% ($P<0.01$) and those in LSD patients by 17.1% ($P<0.05$). Similarly, TGF-β1 levels in SSD patients were elevated by 18.1% ($P<0.05$) relative to LSD patients.

***Table**. Serum Levels of MMPs and TIMPs in Examined Patient Groups are provided in the accompanying table (M±m).*

The studied indicators, ng/ml	Control, n=20	Scleroderma	
		Systemic, n=29	Limited, n=35
MMP-2	203,62±10,91	140,16±7,71*	168,91±6,92*
MMP-7	4,27±0,15	3,12±0,11*Δ	3,65±0,11*
MMP-9	310,61±14,95	212,81±8,82*Δ	260,44±10,68*
TIMP-1	296,54±12,15	422,44±13,51*Δ	357,32±14,43*
TIMP-2	68,33±3,17	92,73±2,19*Δ	77,61±2,02*
TGF-β_1	90,72±4,35	125,42±4,54*Δ	106,22±3,72*

Notes: * - P <0,05 in comparison with the control, Δ - P <0,05 in comparison with LSc.

To evaluate the balance between proteolytic and antiproteolytic activity of serum enzymes in patients with various forms of scleroderma, we analyzed the ratio indices of MMP-9/TIMP-1, MMP-2/TIMP-2, MMP-7/TIMP-1, and MMP-7/TIMP-2. The findings revealed that these ratios were significantly lower in patients with systemic sclerosis (SSD) compared to the control group, showing reductions of 52.4%, 49.3%, 71.4%, and 45.9%, respectively ($P < 0.001$). In patients with localized scleroderma (LSD), the reductions were less pronounced, at 30.5%, 26.8%, 28.6%, and 24.6%, respectively ($P < 0.01$). This indicates a predominance of antiproteolytic activity mediated through regulation by MMP-inhibitory enzymes, a trend that was notably

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more severe in SSD than LSD patients, with differences between the two groups of 31.5%, 30.7% ($P < 0.01$), 60.0% ($P < 0.001$), and 28.3% ($P < 0.01$), respectively, for the observed MMP/TIMP ratios. Additionally, a correlation analysis was conducted to explore relationships between MMPs, TIMPs, and TGF-β1 levels. In SSD patients, TGF-β1 exhibited strong negative correlations with MMP-2 ($r = -0.83$, $P < 0.001$), MMP-7 ($r = -0.81$, $P < 0.001$), and MMP-9 ($r = -0.85$, $P < 0.001$). Conversely, it showed strong positive correlations with TIMP-1 ($r = 0.82$, $P < 0.001$) and TIMP-2 ($r = 0.85$, $P < 0.001$). Similarly, in LSD patients, slightly weaker but still significant correlations were observed with r values of -0.78, -0.76, -0.79, 0.77, and 0.76 ($P < 0.01$) for the respective parameters. These findings demonstrate that in the systemic circulation of scleroderma patients, levels of MMP-2, MMP-9, and MMP-7 are reduced compared to controls, while their inhibitors—TIMP-1 and TIMP-2—show increased activity. This reduction in circulating MMP levels supports the hypothesis that extracellular matrix remodeling is inhibited, contributing to structural and functional abnormalities in the skin and disruption of extracellular matrix metabolism. The lower MMP levels likely reflect their reduced availability in systemic circulation due to upregulated inhibition by TIMP proteins. This imbalance in the MMP/TIMP system appears to be a key mechanism underlying scleroderma development, with a greater degree of imbalance distinguishing SSD from LSD. A notable factor driving the MMP/TIMP imbalance is elevated TGF-β1 levels, as shown by their strong correlations with both MMPs and TIMPs. Higher TGF-β1 levels are associated with increased TIMP-1 and TIMP-2 activity and reduced proteolytic action of MMP-2, MMP-9, and MMP-7. These disruptions in the balance between MMPs and TIMPs offer a theoretical framework for understanding fibrogenesis mechanisms and highlight TGF-β1's regulatory role in extracellular matrix dynamics across different forms of scleroderma.

In conclusion, the imbalance between MMPs and their inhibitors, along with elevated TGF-β1 levels, is instrumental in the pathogenesis of scleroderma. The severity of these disturbances serves as an indicator of disease form, reducing extracellular matrix breakdown and promoting excessive sclerotic processes in the skin. This insight has clinical implications for developing therapeutic approaches aimed at correcting extracellular matrix dysfunction in scleroderma patients.

LITERATURE

1. Antiga E., Quaglino P., Bellandi S. et. al. Regulatory T-cells in the skin lesions and blood of patients with systematic sclerosis and morphoea // Br. J. Dermatol .- 2010. Vol. 162, No. 5.-P.1056-1063.
2. Badea J., Taylor M., Rosenberg A., Foldvari M. Pathogenesis and therapeutical approaches for improved topical treatment in localized scleroderma and systemic sclerosis // Rheumatology.- 2009.- Vol.48, No. 3.-P.213 -221.
3. Brodsky TA, Geltser BI, Nevzorova VA Arterial rehydration and respiratory diseases // Vladivostok: Dal.nauka.- 2018. -S.14-21.
4. Bondar IA, Klimontov VV Matrix metalloproteinases and their inhibitors in the development of kidney fibrosis in diabetes mellitus // Problems of endocrinology .- 2022.- №1.- P.39-44.
5. Bobkova IN, Kozlovskaya LV, Lee OA The role of matrix metalloproteinases in the pathogenesis of kidney diseases // Ter.arch.- 2018.- №6.- P.86-90.
6. Jacob M.P. Extracellular matrix remodeling and matrix metalloproteinases in the vascular wall during aging and pathological conditions. Biomed. Pharmacother.-2023.- Vol.57.-P.195-202.
7. Lee M.P., Sweeney G. Insulin increases gelatinize acclivity in rat glomerular meningeal cell Via ERK-and PI-3 kinase-dependent signaling // Diabet. Obes. Metob.-2016 Vol. 8, No. 3. P.281-288.
8. Mclennan S.V., Kelly D.I., Sohache M. et al. Advanced glytation end products decrease mesangial cell MMP-7: a role in matrix accumulation in diabetic nephropathy? // Kidney Int.- 2017.- Vol.72, No. 4.-P.481-488.
9. Mclenvian S.V., Wang X.Y., Moreno V. Connective tissue growth factor mediates high glucose effects on the matrix of inhibition of inhibitor nephropathy // Endocrinology .- 2014.- Vol.145.- P.5646-5655.
10. Taylor A. W. Review of the activation of TGF-beta in immunity. // J. Leukoc. Biol .- 2009. Vol.85, No. 1.-P.29-33.
11. Volukhin VA, Muraden NL, Katunina O.R. About the role of profibroznyh cytokines in the pathogenesis of localized scleroderma // Herald of Dermatology and Venereology .- 2022.- №1 .- P.28-33.
12. Yamamato T. Scleroderma-pathophysiology //Eur.J. Dermatol .- 2019.- Vol.19, No. 1.-P.14-24.