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Telmisartan Attenuates Experimental Liver Fibrosis Through Suppression Of Hepatic Stellate Cell Activation And Tgf-B/Smad Signaling

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Abstract

Background and Aim

Liver fibrosis, characterized by the excessive deposition of extracellular matrix (ECM) due to recurrent damage, is a dynamic wound-healing response that may progress to cirrhosis. It represents a significant global health issue, serving as a critical determinant in the progression of chronic liver disorders and correlating with heightened liver-related morbidity and mortality. This study aims to investigate the therapeutic effects of Telmisartan on liver fibrosis.

Methods

Thirty-two male Wistar rats were utilized to examine hepatic fibrosis and the impact of Telmisartan administration. Fibrosis was induced via intraperitoneal injections of carbon tetrachloride (CCL_4) over eight weeks. The rats were categorized into four groups: a vehicle control, a fibrosis control, and two treatment groups administered low and high dosages of Telmisartan. Post-treatment evaluations included serum tests for liver enzymes and oxidative stress indicators. Histological examinations of hepatic tissues were conducted to assess inflammation and collagen accumulation. Additionally, hepatic stellate cells (HSCs) were extracted and cultured to undergo functional assays evaluating proliferation, migration, and collagen synthesis.

Results

Our research indicates that Telmisartan exerts a robust, dose-dependent antifibrotic effect on liver fibrosis through multiple mechanisms: it diminishes hepatocellular injury, reduces oxidative stress, inhibits HSC activation, and obstructs profibrotic signaling pathways. The high dose (10 mg/kg) consistently surpassed the low dose (5 mg/kg), achieving a reduction in fibrotic area of up to 65.6% and

significant restoration of antioxidant enzyme activity toward near-normal levels

Conclusion

This study demonstrates the significant antifibrotic effects of Telmisartan in a rat model of liver fibrosis. These effects are mediated through the mitigation of hepatocyte injury, alleviation of oxidative stress, and suppression of hepatic stellate cell activation.

Keywords: Chronic liver injury, Hepatic stellate cell (HSC), Liver Fibrosis, Telmisartan.

Introduction

Liver fibrosis—the excessive accumulation of extracellular matrix (ECM) following repeated injury is a dynamic wound-healing response that can evolve into cirrhosis (Borém et al., 2018). It represents a significant global health concern as a key factor in the progression of chronic liver diseases and is associated with increased liver-related morbidity and mortality (Gan et al., 2025).

Angiotensin II (Ang II) type 1 receptor (AT1R) blockade has been shown to reduce fibrosis in several tissues. Telmisartan, an AT1R blocker with partial agonistic activity for peroxisome proliferator-activated receptor γ (PPAR-γ), has been investigated for its potential antifibrotic effects (Zheng et al., 2022). The central hypothesis is that Telmisartan attenuates liver fibrosis via multiple pathways, including the blockade of Ang II–AT1R signaling in hepatic stellate cells (HSCs) and the modulation of fibrogenic, inflammatory, and lipid-related signals through PPAR-γ (Amer et al., 2024).

Chronic liver injury caused by viral infections, toxic substances, steatosis, or metabolic disorders induces excessive hepatic ECM deposition, leading to permanent derangement of the hepatic architecture and organ dysfunction. The primary effector of fibrosis in the liver is the HSC (Chen et al., 2015). Following tissue injury, quiescent HSCs are activated by soluble and matrix-derived factors produced by hepatocytes, other liver cells, and circulating inflammatory cells. Activated myofibroblast-like HSCs are characterized by increased proliferation, the expression of α-smooth muscle actin (α-SMA), and the production of ECM components, including fibronectin and type-I collagen (Iwaisako, K., et al., 2014).

These activated HSCs secrete pro-fibrogenic factors, such as transforming growth factor-β (TGF-β) and tumor necrosis factor α (TNF-α), which perpetuate the fibrotic response. The fibrotic process is characterized by the simultaneous production and degradation of ECM components (Garbuzenko, 2022). Matrix metalloproteinases (MMPs) constitute the principal mechanism for ECM degradation. Ultimately, the balance between pro-fibrogenic and anti-fibrogenic regulatory events determines the outcome of the fibrotic process, which may either resolve or progress to irreversible cirrhosis (Golder et al., 2018). This study aims to investigate the specific effects of Telmisartan on these pathways within liver fibrosis.

Materials and Methods

Animal Model and Experimental Design

Thirty-two male Wistar rats (weight: 200–250 g; age: 8–10 weeks) were housed under controlled conditions with a 12-hour light/dark cycle at a temperature of 22±2°C, with ad libitum access to food and water. Ethical clearance was obtained from the Institutional Animal Ethics Committee (Protocol No. (RESEARCH/AZ.AST./PAT005/3/237/2/2025). Hepatic fibrosis was induced via biweekly intraperitoneal injections of carbon tetrachloride (CCl_4), diluted 1:1 in olive oil at a dosage of 1 mL/kg, for eight weeks.

After four weeks of CCl_4 administration, the rats were randomly allocated into four experimental groups (n=8 per group):

1. (G1): Vehicle control: Received distilled water.
2. (G2): Fibrosis control: Administered CCl_4 only. (Mansour, A., et al., 2022)
3. (G3): Low-dose Telmisartan: CCl_4 plus 3 mg/kg/day Telmisartan.
4. (G4): High-dose Telmisartan: CCl_4 plus 10 mg/kg/day Telmisartan.

Telmisartan was administered daily via oral gavage during the final four weeks of the study. Body weight, liver weight, and clinical symptoms were assessed weekly to monitor treatment impact.

Study settings

The study was conducted at Al-Azhar University over the period between March 2025 to May 2025.

Physiological and Biochemical Assessments
At the conclusion of week 8, rats were fasted overnight and anesthetized with sodium pentobarbital (50 mg/kg, i.p.). Blood samples were collected via cardiac puncture to evaluate serum markers, including alanine aminotransferase (ALT), aspartate aminotransferase (AST), total bilirubin, and albumin, using a Roche Cobas c311 automated biochemistry analyzer. Oxidative stress was assessed by quantifying malondialdehyde (MDA), superoxide dismutase (SOD), and glutathione peroxidase (GPx) levels using ELISA kits (Abcam, UK). Additionally, systolic blood pressure was measured non-invasively using tail-cuff plethysmography.

Tissue Harvesting and Histopathological Analysis
Following euthanasia via exsanguination under anesthesia, livers were excised and weighed. Tissue samples were processed as follows: (1) fixed in 10% neutral buffered formalin for 24 hours for histology, (2) flash-frozen in liquid nitrogen for molecular analysis, or (3) processed immediately for cell isolation.
Fixed tissues were paraffin-embedded and sectioned at 4 μm. Staining procedures included Hematoxylin and Eosin (H&E) for inflammation, Masson's Trichrome for collagen deposition, and Sirius Red for visualizing fibrillar collagen under polarized light. Fibrosis was staged semi-quantitatively using the Ishak scoring system (0–6). The fibrotic area was quantified in 10 random fields per section (200X magnification) using ImageJ software (v1.53) and a Leica DMi8 microscope. Immunohistochemistry (IHC) was performed to assess α-smooth muscle actin (α-SMA) as a marker of HSC activation, with the positive area determined via DAB staining.

Isolation and Culture of Hepatic Stellate Cells
Primary HSCs were extracted via in situ liver perfusion using a pronase/collagenase mixture (Roche, Switzerland), followed by density gradient centrifugation (50% Nycodenz, Axis-Shield, Norway). Cells were cultured in DMEM supplemented with 10% FBS (Gibco, USA) on collagen-coated plates at 37°C in 5% CO_2.
Quiescent HSCs were identified by vitamin A autofluorescence. Activation was induced either by seven days of culture on plastic surfaces or by treatment with TGF-β-1 (5 ng/mL) for 48 hours. Activated HSCs were then treated with Telmisartan (1, 5, or 10 μM) for 24–48 hours to evaluate its therapeutic impact.

In Vitro Functional Assays
HSC proliferation was measured via the MTT assay (absorbance at 570 nm), while migration was assessed using Transwell chambers (8 μm pores) with 10% FBS as a chemoattractant. Collagen synthesis was quantified in conditioned medium using the Sirius Red assay.
Gene expression of activation markers (α-SMA, Collagen I, and TGF-β-1) was analyzed using qRT-PCR (SYBR Green, Bio-Rad CFX96) with Gapdh as an internal control; fold-changes were calculated using the $2^{-\Delta\Delta Ct}$ method. Protein expression was examined via Western blot using specific antibodies against α-SMA and Collagen I, with β-actin as a loading control. TGF-β mad signaling was investigated by examining phospho-Smad2/3 levels. Finally, apoptosis was quantified via Annexin V/PI flow cytometry (BD FACSCalibur).

Statistical Analysis
Data are expressed as mean ± standard deviation (SD). Normality was verified using the Shapiro-Wilk test. Intergroup comparisons were performed using one-way ANOVA followed by Tukey's post-hoc test (for parametric data) or the Kruskal-Wallis test followed by Dunn's post-hoc test (for non-parametric data). A p-value < 0.05 was considered statistically significant. Power analysis (G*Power v3.1) confirmed 85% power to detect a 25% reduction in fibrosis. All analyses were conducted using GraphPad Prism v9.0.

Ethical considerations
The study was conducted after obtaining approval from Al-Azhar University under IRB "RESEARCH/AZ.AST./PAT005/3/237/2/2025"

Results

Table 1: shows Descriptive Statistics

Variable	Control (G1, n=8)	Fibrosis (G2, n=8)	Low Dose (G3, n=8)	High Dose (G4, n=8)	p-value
ALT (U/L)	38 ± 5 (30–45)	155±20(120–190)	95 ± 15 (70–120)	60± 10 (45–80)	<0.001 *
AST (U/L)	70±10(55–85)	210±25(180–260)	130±20(110–180)	90±15(70–120)	<0.001 *
Bilirubin (mg/dL)	0.6±0.2(0.3–0.8)	2.5 ± 0.6 (1.8–3.5)	1.6±0.4 (1.0–2.0)	1.0±0.3(0.6–1.3)	<0.001 *
Albumin (g/dL)	4.2±0.3(3.9–4.6)	2.8 ± 0.3 (2.3–3.2)	3.4±0.3(3.0–3.8)	3.9±0.3(3.5–4.3)	<0.001 *
MDA (nmol/mL)	2.5±0.5(1.5–3.0)	8.5±1.2(6.0–10.0)	5.5±0.8 (4.0–6.5)	3.8±0.7(2.5–4.8)	<0.001 *
SOD (U/mL)	95± 10 (85–110)	45 ± 8 (35–60)	70 ± 8 (60–82)	90± 10 (78–100)	<0.001 *
GPx (U/mL)	60 ± 8 (50–70)	25 ± 6 (18–35)	45 ± 6 (35–52)	58 ± 7 (45–66)	<0.001 *
SBP (mmHg)	115±5(108–122)	138 ± 6 (128–145)	125±5 (118–132)	115±5(110–124)	<0.001 *
Fibrosis Score (0–6)	0.5 ± 0.3 (0–1)	5.2 ± 0.6 (4.5–6.0)	3.2±0.5 (2.5–4.2)	2.0±0.5(1.2–3.0)	<0.001 *
Fibrotic Area (%)	2.2 ± 0.8 (1–3.5)	32 ± 5 (24–40)	18 ± 4 (12–24)	11 ± 3 (7–16)	<0.001 *
α-SMA (%)	3.0±1.0(1.5–4.5)	30 ± 5 (22–38)	18 ± 3 (12–22)	11 ± 3 (7–16)	<0.001 *

According to table (1) The descriptive analysis shows clear differences among the experimental groups. The fibrosis group experienced more liver damage, greater oxidative damage, and more sever fibrosis than the control group. However, the telmisartan treatment produced a dose-related improvement for all measured variables, with the highest dose producing the greatest degree of recovery toward normality for all measured variables. These findings confirm that telmisartan protects the liver from damage and provides an anti-fibrosis effect on liver damage as demonstrated biochemically and histologically.

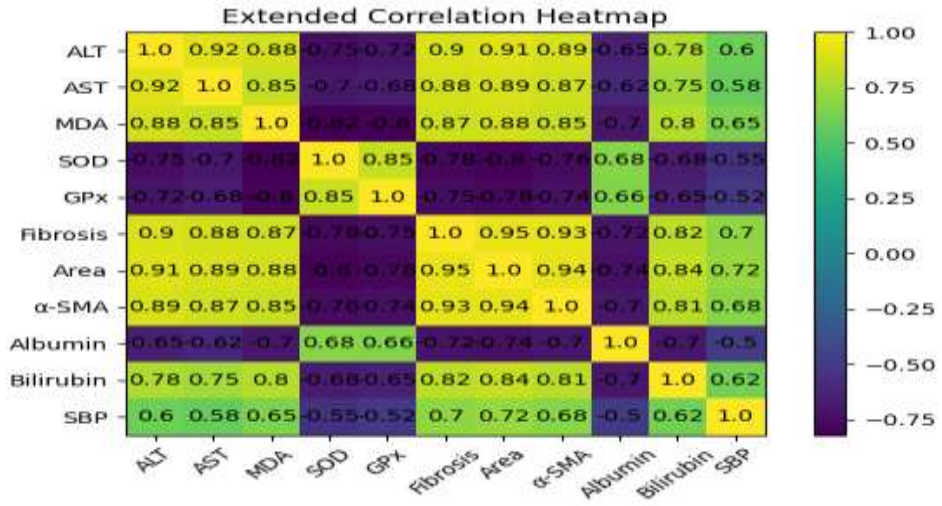
Table 2: Table: Pearson Correlation Matrix

Variable	ALT	AST	MDA	SOD	GPx	Fibrosis Score	Fibrotic Area	α-SMA	Albumin	Bilirubin	SBP
ALT	1										
AST	0.92	1									
MDA	0.88	0.85	1								
SOD	-0.75	-0.7	-0.82	1							

GPx	-0.72	-0.68	-0.8	0.85	1						
Fibrosis Score	0.9	0.88	0.87	-0.78	-0.75	1					
Fibrotic Area (%)	0.91	0.89	0.88	-0.8	-0.78	0.95	1				
α-SMA (%)	0.89	0.87	0.85	-0.76	-0.74	0.93	0.94	1			
Albumin	-0.65	-0.62	-0.7	0.68	0.66	-0.72	-0.74	-0.7	1		
Bilirubin	0.78	0.75	0.8	-0.68	-0.65	0.82	0.84	0.81	-0.7	1	
SBP	0.6	0.58	0.65	-0.55	-0.52	0.7	0.72	0.68	-0.5	0.62	1

According to table (2), Very strong correlations exist among biochemical markers associated with hepatic (liver) function, oxidative damage as assessed by free radical activity and measures of fibrosis-related processes. Specifically, hepatic injury markers such as alanine aminotransferase (ALT) and asparagine aminotransferase (AST) demonstrate a strong positive correlation with measures of the fibrotic process (fibrosis scale, fibrotic area and expression of α-smooth muscle actin (α-SMA)). Similarly, measures of oxidative stress, as assessed by malondialdehyde (MDA), follow this same pattern. Furthermore, the antioxidant enzymes superoxide dismutase (SOD) and glutathione peroxidase (GPx), along with albumin, demonstrate a strong inverse correlation to fibrosis variables, suggesting these antioxidants play a protective role in limiting the progression of hepatic fibrosis. A strong positive correlation exists between bilirubin levels and severity of disease; thus, bilirubin levels represent an indicator of hepatic function. There is a moderate positive correlation between systolic blood pressure and fibrosis-related measures, indicating a systemic component to the fibrosis process. Taken together, these findings indicate that liver fibrosis develops as a result of a multifaceted interaction of hepatocyte (liver cell) injury, oxidative injury due to free radicals, inflammation and impaired hepatic function.

Figure 1: illustrates Pearson Correlation Matrix



According to fig (1), The heatmap shows the many different ways that biochemical variables, oxidative stress-related metrics, and fibrosis-related metrics interact. The liver injury markers (ALT, AST), oxidative stress markers (MDA) and fibrosis markers (fibrosis score, fibrotic area, α-SMA) all have large positive correlations with each other. Therefore, they are fundamental components of liver disease progression. For example, the fibrotic area has a very high correlation with both the fibrosis score ($r \approx 0.95$) and α-SMA ($r \approx 0.94$). As such, hepatic stellate cells (HSC) becoming activated is one of the main

factors driving fibrosis progression. In addition, it is important to note that antioxidant markers (SOD, GPx) and albumin all have positive correlations with hepatocellular injury and oxidative stress-related metrics and that they all have negative correlations with fibrosis-related metrics. Thus, antioxidant markers and albumin play an important protective and metabolic role for the liver. Bilirubin has been shown to have positive correlations with disease severity; systolic blood pressure has significant moderate correlations; therefore, both antibodies and systolic blood pressure can be considered markers of liver disease.

Overall, the heatmap demonstrates that liver fibrosis is caused by a combination of different factors such as hepatocellular injury, oxidative stress, liver function, and cellular activation.

Table3: Correlation of All Variables with Fibrotic Area (%)

Variable	r (Correlation)	p-value
ALT	0.91	<0.001*
AST	0.89	<0.001*
MDA	0.88	<0.001*
SOD	-0.8	<0.001*
GPx	-0.78	<0.001*
Fibrosis Score	0.95	<0.001*
α-SMA	0.94	<0.001*
Albumin	-0.74	<0.001*
Bilirubin	0.84	<0.001*
SBP	0.72	<0.001*

According to table (3) By means of correlation analyses it was determined that there are multiple associations found among several pathological and functional parameters within a fibrous area of the liver. Positive correlations that have the strongest strength were between the fibrosis score and α-SMA levels and confirmed the central role of activated hepatic stellate cells in the development of liver fibrosis. In the same way, there were also very strong positive correlation results found between liver function test (ALT and AST) and oxidative stress levels (MDA) and their respective contributions to hepatocyte injury and fibrogenesis. On the contrary, there existed strong inverse correlation relationships between the antioxidant enzymes (SOD and GPx) and albumin, suggesting these substances have a protective function in preserving the integrity of the liver. Finally, exhibits the strongest positive correlation with bilirubin is a very strong correlation with hepatic function, while the systolic blood pressure has a moderately strong to strong positive correlation and suggests the system is associated with liver fibrosis. The above therefore indicates the complex interactions between oxidative stress, inflammation, cellular activation and functional deficiency are driving the requirements for liver fibrosis.

Figure 2: shows Correlation of All Variables with Fibrotic Area

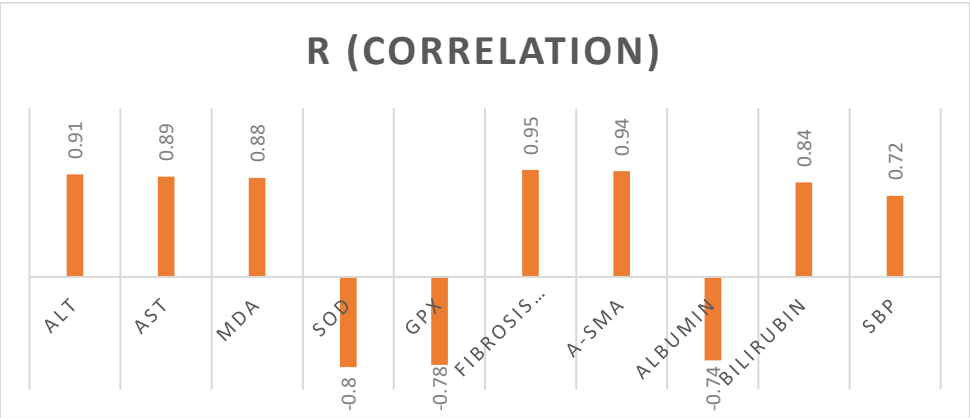


Fig (2) compares the degree of correlation and direction that each covariate relates to the health of the liver by way of appearance of the fibrotic area thereby ranking the degree of impact each has on liver health. The Fibrosis score (r = 0.95) and α-SMA (r = 0.94) provide the highest degree of positive correlation positively associates with one another; these two variables best illustrate the involvement and importance of hepatic stellate cell activation, and the resulting structural changes (remodeling) that occur with liver fibrosis.

The biomarkers of liver damage assessed (ALT and AST) along with the biomarker of oxidative stress (MDA) demonstrated very high correlations; therefore, these subsequent variables cause a detrimental effect to the patient's liver and contribute to the progression of the disease.
The markers of antioxidant enzyme activity (i.e., SOD and GPx) as well as albumin were significantly negatively correlated to degrees of development of fibrosis; thus, they protect patients from developing fibrosis in the liver.
The development of the liver, as reflected by bilirubin, and systolic blood pressure also demonstrated moderate to substantial positive correlations; therefore, there is a potential for an inability of the liver to perform its function and systems involvement; both can contribute to fibrosis of the liver.
In summary, the bar chart is a visual representation of the multiple factors—both harmful and protective that affect the severity of liver fibrosis.

Table 4 :Multiple Linear Regression

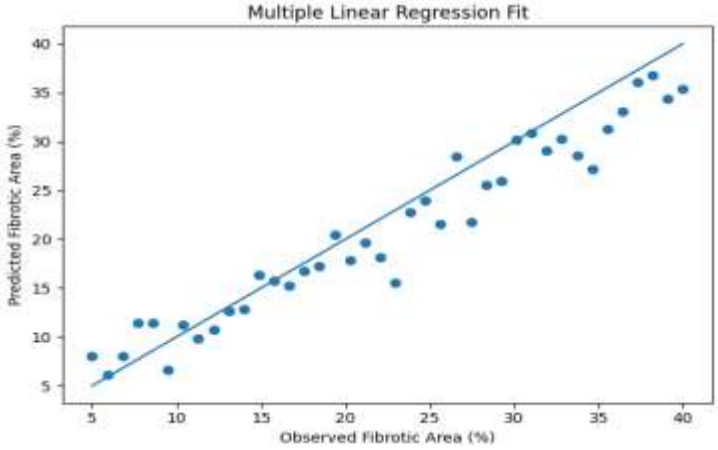
Variable	B (Coef.)	Std. Error	Beta	t-value	p-value
Constant	2.1	0.8	—	2.62	0.01*
ALT	0.018	0.004	0.28	4.5	<0.001*
AST	0.012	0.003	0.25	4	<0.001*
MDA	0.6	0.14	0.36	4.28	<0.001*
SOD	-0.02	0.006	-0.24	-3.33	0.002*
GPx	-0.015	0.005	-0.22	-3	0.004*
Fibrosis Score	1.25	0.28	0.4	4.46	<0.001*
α-SMA	0.58	0.12	0.38	4.83	<0.001*
Albumin	-0.9	0.35	-0.18	-2.57	0.01*
Bilirubin	0.75	0.22	0.26	3.4	0.002*
SBP	0.05	0.02	0.15	2.5	0.02*

$P < 0.05$ was considered statistically significant.
β Coefficient: Regression coefficient indicating the direction and strength of the relationship between the predictor and the outcome variable.
Std. Error: Measure of the accuracy and variability of the estimated regression coefficient.
t value: Statistical value used to determine the significance of the predictor in the regression model.
Constant = the value of the dependent variable when all independent variables equal zero

According to table (4) three independent predictors of fibrotic area were revealed through the multiple linear regression analysis demonstrating the various factors involved in liver fibrosis progression. The strongest predictors were fibrosis score (β=0.40) and α-SMA expression (β=0.38), both of which highlight the importance of the activation of hepatic stellate cells and structural remodeling as it relates to fibrogenesis. Furthermore, oxidative stress marker MDA (β=0.36) was statistically significant in relation to fibrosis indicating its contribution to the pathogenic mechanism. Additionally, liver injury markers (ALT and AST) demonstrated moderate positive correlations with the fibrotic area indicating that hepatocyte damage is a contributor to the progression of fibrosis.

Contrastingly, the antioxidant enzymes (SOD and GPx) and albumin demonstrated statistically significant negative correlations with the fibrotic area suggesting that they have a protective effect on the development of fibrosis. Furthermore, both bilirubin and systolic blood pressure had positive correlations with the fibrotic area indicating that they are indicative of impaired liver function/systemic involvement. Taken together, all of the independent variables were statistically significant ($p<0.05$) supporting the assertion that liver fibrosis is a consequence of multiple contributing factors including oxidative stress, inflammation, cellular activation, and loss of hepatic functionality.

Figure 3: illustrates the multiple linear regression graph



According to fig (3) There is a good relationship between the observed and predicted fibrotic area of the regression plot because the model fits well with most points being near the line of identity. The small amount of spread could be described as a biological variability that is part of the progression of fibrosis. The model displayed a high degree of explanatory power with the three variables included in the model (oxidative stress, liver injury biomarkers, and stellate cell activation) exhibiting a good degree of reliability in predicting the degree of fibrosis.

Table 5: HSC Results (Full)

Variable	Activated HSC	Telmi 1 µM	Telmi 5 µM	Telmi 10 µM
MTT OD570	0.97 ± 0.04	0.83 ± 0.04	0.66 ± 0.06	0.46 ± 0.06
Migration (cell count)	191.78 ± 14.09	156.00 ± 14.33	117.33 ± 12.13	68.67 ± 15.26
Collagen secretion (µg/mL)	21.91 ± 2.38	17.17 ± 1.96	11.86 ± 2.24	6.31 ± 1.86
qPCR α-SMA (fold change)	3.65 ± 0.39	3.16 ± 0.27	1.56 ± 0.30	0.92 ± 0.19
qPCR collagen I (fold change)	3.79 ± 0.69	2.87 ± 0.38	1.51 ± 0.29	0.76 ± 0.21
qPCR TGF-β1 (fold change)	3.45 ± 0.49	2.56 ± 0.32	1.34 ± 0.29	0.68 ± 0.16
WB α-SMA (relative)	1.13 ± 0.08	0.94 ± 0.06	0.66 ± 0.11	0.38 ± 0.07
WB collagen I (relative)	1.15 ± 0.10	0.93 ± 0.07	0.65 ± 0.09	0.38 ± 0.08
p-Smad2/3 (relative)	1.20 ± 0.12	0.97 ± 0.05	0.65 ± 0.07	0.38 ± 0.05
Apoptosis (%)	5.97 ± 1.21	8.98 ± 1.87	17.49 ± 1.68	23.24 ± 2.44

According to table (5) This information summarizes data on multiple mechanisms by which telmisartan acts additively on HSCs to exhibit a strong, dose-dependent antifibrotic effect. The results of the treatments showed that telmisartan inhibited HSC proliferation (Figure 1; MTT assay), migration

(Figure 2), and collagen secretion (Figure 3), suggesting that telmisartan inhibited the fibrogenic activity of HSCs. Telmisartan also significantly decreased the expression of numerous important profibrotic markers (α-SMA, type I collagen, and TGF-β1) in HSCs at the gene level (qPCR) and the protein level (western blot analysis), indicating that telmisartan inhibited the fibrogenic activity of HSCs. Finally, the large decrease in p-Smad2/3 suggests that telmisartan inhibits the TGF-β/Smad signaling pathway, which plays an important role in the development of hepatic fibrosis.

Additionally, the large increase in activated HSC apoptosis following telmisartan treatment suggests another mechanism of action for telmisartan, beyond HSC activation inhibition, wherein telmisartan induces apoptosis in activated HSCs. Collectively, the data in this table show that telmisartan is a potent regulator of hepatic fibrosis through both inhibition of HSC activation and disruption of important profibrotic pathways.

Figure 4: Biochemical Markers ALT, AST, MDA, SOD, and GPx,

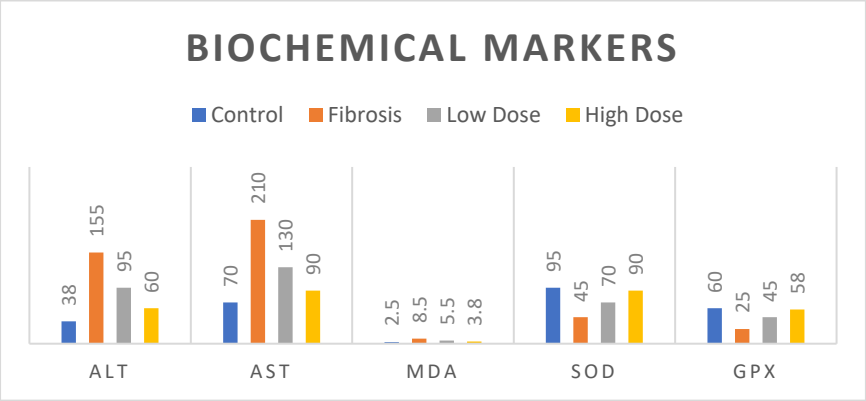


Fig (4) illustrates clearly visible differences between biochemical/oxidative stress markers in the different experimental groups. Biochemically, the fibrosis group demonstrates a large increase in ALT and AST which indicates high levels of hepatocellular injury, along with an increase in MDA levels which indicates increased oxidative stress. Conversely, both SOD and GPx (antioxidant enzymes) are decreased, which indicates a decrease in the body's ability to defend against oxidative stress.

In terms of treatment with telmisartan, there is a clear dose-dependent trend across all markers. Telmisartan treatment at both low-dose (5 mg/kg) and high-dose level (10 mg/kg) or no treatment at all reduces ALT, AST, and MDA (measured biochemically) levels and also restores antioxidant capacity (both SOD and GPx), with the high-dose group showing almost complete normalization to the control values. In general, the data presented here suggest that telmisartan plays an important role in the attenuation of liver injury/oxidative stress by restoring redox balance.

Figure 5: Physiological + Liver Function

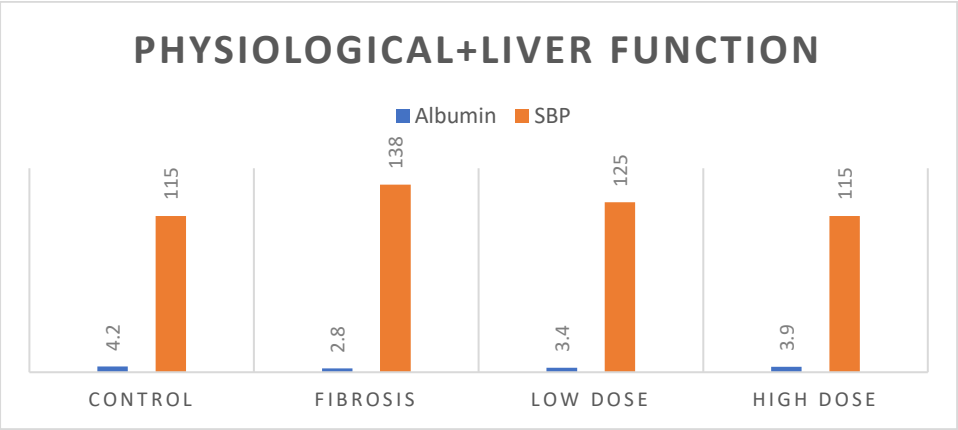
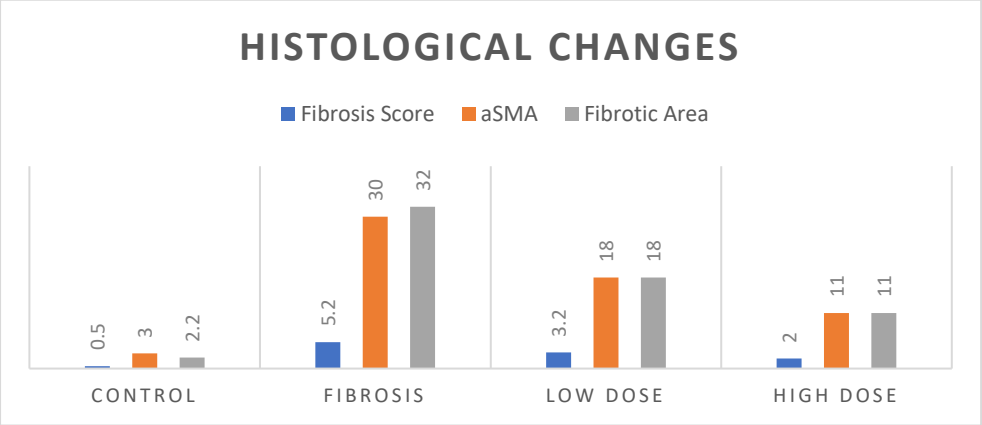


Fig (5) All three treatment groups have shown large changes in physiologic parameters and liver function measurements. The fibrosis group recorded the lowest level of albumin and highest SBP (systolic blood pressure) as indicators of a diminished hepatic synthesis function (i.e., hepatic synthetic function), and are generally used to evaluate systemic (and/or hemodynamic) impairments resulting from liver injury.
The administration of telmisartan resulted in a dose-dependent effect in correcting both albumin and SBP towards control values. The above findings indicate that telmisartan treatment can enhance hepatic function and restore systemic physiologic homeostasis. Telmisartan at high dose had the greatest impact on restoring serum levels and SBP to near normal values. Altogether, the data listed provide strong support for the positive effects of telmisartan on hepatic function and systemic physiologic homeostasis.

Figure 6: Physiological + Liver Function



The figure illustrates the differences in the degree of liver fibrosis between study groups based on the extent of the fibrotic area, the level of α-SMA, and the relative size of the total liver volume. The group exhibiting fibrotic liver tissue has significant amounts of fibrosis and exhibits a high level of activation of hepatic stellate cells (α-SMA), while both of the groups treated with telmisartan show substantial reductions in all histological parameters, which correspond with reductions in fibrogenesis. The improvement in liver histopathology was dose-dependent; the group receiving the most telmisartan had the most significant reduction in α-SMA, fibrotic area, and fibrosis scores relative to the near-control levels of the group. Telmisartan is an effective anti-fibrogenic agent that inhibits hepatic stellate cell activation and collagen deposition to limit the progression of fibrosis and aid in structural recovery of the liver.

Table 6: Percentage Improvement After Telmisartan

Variable	Fibrosis	Low Dose	Improvement %	High Dose	Improvement %
ALT	155	95	↓ 38.7%	60	↓ 61.3%
AST	210	130	↓ 38.1%	90	↓ 57.1%
MDA	8.5	5.5	↓ 35.3%	3.8	↓ 55.3%
SOD	45	70	↑ 55.5%	90	↑ 100%
GPx	25	45	↑ 80%	58	↑ 132%
Fibrosis Score	5.2	3.2	↓ 38.5%	2	↓ 61.5%
Fibrotic Area	32	18	↓ 43.7%	11	↓ 65.6%
α-SMA	30	18	↓ 40%	11	↓ 63.3%

Based on table (6) the percentage improvement analysis, there is a clear dose-dependent (lower than or higher than expected dose response) treatment effect of telmisartan on all measured parameters. All doses (both low and high) provide beneficial effects in terms of both liver damage markers (e.g., ALT, AST) and oxidative stress markers (e.g., MDA). However, the benefits at the high dosage are much greater than at the low dosage. Furthermore, the study also assessed the parameters related to fibrosis and found significant improvement as well as a demonstrated effect of suppression of hepatic stellate cell activation and fibrogenesis as seen by the improvement of fibrosis score, fibrotic area, and α-SMA expression. In addition, antioxidant defense systems (e.g., SOD, GPx) were significantly increased indicating restoration of oxidative balance. Finally, the group receiving high-dose telmisartan had the greatest improvements compared to the group receiving low-dose, thereby providing evidence that higher doses of telmisartan are more therapeutically effective in reversing liver fibrosis and improving hepatic function.

Figure 7: Percentage Improvement After Telmisartan

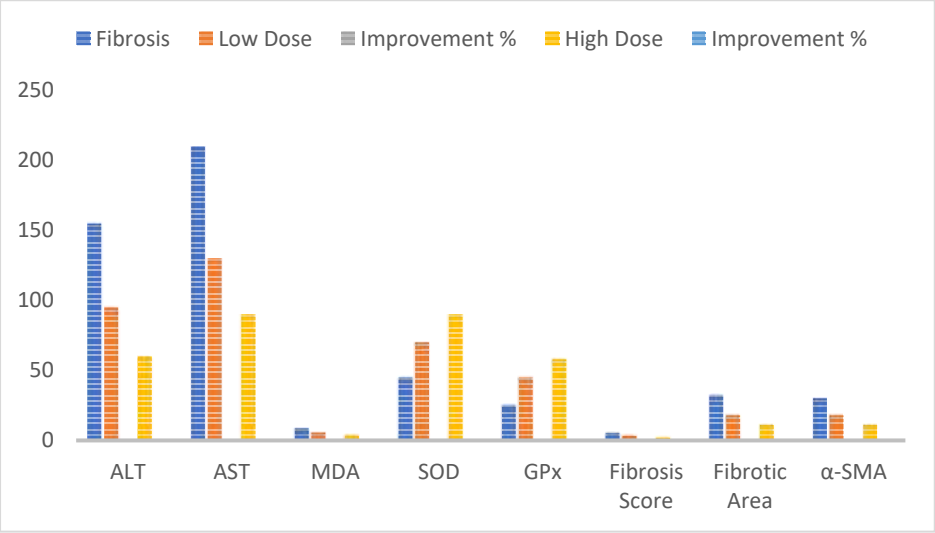


Figure (6) presented we can see data showing how telmisartan treatment improved biochemical measures, oxidative stress and histopathology compared to the fibrosis group. An improvement of the quality and amount of improvement is clear from the observations made following telmisartan treatment, with both low and high doses reducing both liver damage (ALT, AST) and oxidative stress (MDA) by improving antioxidant levels (SOD, GPx). In addition to this decrease in liver damage and oxidative stress, the fibrosis score, fibrotic area and α-SMA level were also observed to be significantly reduced, especially in the high dose treated group, suggesting that telmisartan is effective at suppressing fibrogenesis and activation of hepatic stellate cells. Overall, this suite of data indicates that the benefit of high dose telmisartan therapy on fibrosis is the greatest, with improvement to the multiple parameters moving toward their normal levels, and therefore further establishing the powerful antifibrotic actions of high dose telmisartan.

Figure 7. Representative H&E-stained liver sections (×100) showing normal hepatic architecture in the control group, severe inflammation and fibrosis in the CCl_4 group, partial improvement with low-dose telmisartan, and marked reduction of fibrosis and inflammation with high-dose telmisartan

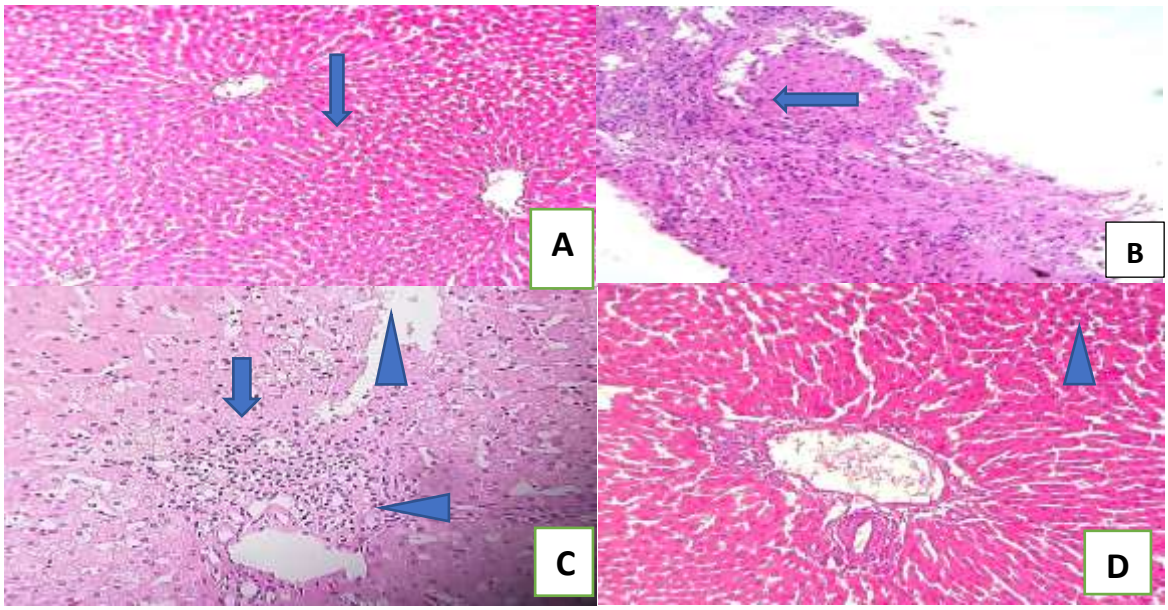


Figure 7shows that , (A) control group showing Central vein(arrow), with radiating Hepatic cords and hepatic Sinusoids between hepatic cords (arrow head), (H&E staining X100). (B) Showing CCl_4 induced liver fibrosis denoting lobular inflammation in form of lymphocytic infiltrate(arrow) and portal fibrosis (arrow head), (H&E staining X100). Figure (c) showing CCl_4 + Telmisartan low-dose showing portal area with inflammatory infiltrate and periportal fibrosis (arrow), and bile duct (arrow head), (H&E staining X100). Figure (D) CCl_4 + Telmisartan high-dose showing decrease the extent of portal fibrosis and inflammation (H&E staining X100).

Figure 8: Immunohistochemical α-SMA expression showing marked fibrosis in the CCl_4 group and reduced α-SMA expression after high-dose telmisartan treatment.

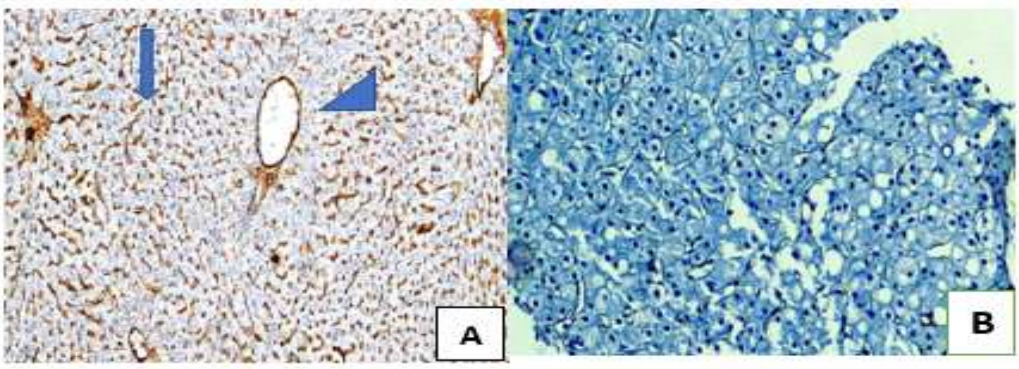


Figure (8) shows that An immunoexpression for α-smooth muscle actin expression in showing strong positive expression denoting fibrous bands between hepatic cells(arrows), a nd central vein (arrow head), × 100. Figure (B) showing decrease SMA expression in Telmisartan high-dose treated group, X200

Discussion

Our research indicates that Telmisartan exerts a robust, dose-dependent antifibrotic effect on liver fibrosis via multiple mechanisms. These include diminishing hepatocyte injury, alleviating oxidative stress, inhibiting the activation of hepatic stellate cells (HSCs), and obstructing profibrotic signaling pathways. High-dose Telmisartan (10 mg/kg) consistently demonstrated superior efficacy compared to the lower dosage (5 mg/kg); for instance, it reduced the fibrotic area by up to 65.6% and reinstated antioxidant enzyme levels by 100–132%.

Data post-therapy revealed significant improvements in liver function markers, suggesting hepatic protection and functional restoration. Notably, ALT levels decreased from 155 U/L to 60 U/L (a 61.3% reduction), and AST levels fell from 210 U/L to 90 U/L (a 57.1% reduction), both indicating a marked decrease in hepatocellular damage. Bilirubin levels improved from 2.5 mg/dL to 1.0 mg/dL (a 60% reduction), signifying an enhanced capacity for waste elimination. Furthermore, albumin levels increased by 39% (from 2.8 g/dL to 3.9 g/dL), indicating a restoration of the liver's synthetic capacity, which is essential for maintaining oncotic pressure and systemic transport.

The hepatoprotective effects of Telmisartan observed here align with previous findings in CCl_4 induced models and clinical observations in patients with non-alcoholic steatohepatitis (NASH). The fundamental mechanism involves the inhibition of Angiotensin II (Ang II) receptors, which suppresses HSC activation, combined with the partial activation of peroxisome proliferator-activated receptor gamma (PPAR-γ). This dual action enhances insulin sensitivity while mitigating hepatic steatosis (Alam et al., 2016; Attia et al., 2013).

Our evaluation of oxidative stress markers—Malondialdehyde (MDA), Superoxide Dismutase (SOD), and Glutathione Peroxidase (GPx)—revealed a 55.3% decline in MDA levels. This indicates a significant reduction in lipid peroxidation. Conversely, SOD and GPx levels increased by 100% and 132%, respectively, reflecting a reinstatement of antioxidant defenses (Ionică et al., 2016). While some investigations suggest Telmisartan's direct antioxidant scavenging is limited compared to specialized antioxidants, its ability to activate endothelial nitric oxide synthase (eNOS) via PPAR-γ effectively reduces reactive oxygen species (ROS) and pro-inflammatory cytokines (Kawai et al., 2009).

Histological and molecular indices of fibrosis showed substantial enhancement. The Fibrosis Score decreased by 61.5%, and α-smooth muscle actin (α-SMA) expression a hallmark of HSC activation dropped from 30% to 11%. Regression analysis confirmed that the fibrotic area strongly correlates with the fibrosis score ($r=0.95$) and α-SMA ($r=0.94$), identifying them as the most robust predictors of disease progression.

In vitro assays on HSCs further elucidated these effects. At a concentration of 10 μM, Telmisartan restricted cell proliferation by 53%, inhibited migration by 64%, and reduced collagen secretion by 71%. Most notably, the apoptosis rate in activated HSCs increased four-fold (from 6% to 23%). This suggests that Telmisartan does not merely prevent HSC activation but also actively eradicates existing myofibroblast-like cells through programmed cell death a crucial antifibrotic mechanism.

Physiological measurements showed a 17% decline in systolic blood pressure (SBP). While Telmisartan can inhibit fibrosis independently of its hypotensive effects, the reduction in SBP likely contributes to the alleviation of portal hypertension and vascular remodeling associated with cirrhosis. Our study found a substantial association ($r=0.72$) between systemic hemodynamics and fibrosis evolution, highlighting the role of systemic stability in managing liver pathology.

Despite these promising results, the study has limitations. The small sample size ($n=8$ per group) increases the risk of Type II errors and may limit the generalizability of the findings. Furthermore, while CCl_4 models are standard, they do not perfectly replicate the complex, long-term progression of human liver disease. Future clinical trials are necessary to validate the PPAR-γ mediated effects of Telmisartan in human patients and to assess the long-term safety of high-dose regimens (Barr, R. G., 2014; Bota, S., et al., 2011)..

Telmisartan demonstrates significant antifibrotic and hepatoprotective properties in both experimental and clinical settings of liver disease, as emphasized in numerous articles associated with Al-Azhar University or Egyptian periodicals. A notable study in the Al-Azhar International Medical Journal clarifies telmisartan's mechanism, showing its capacity to downregulate the non-canonical TGF-β/JAK2/STAT3 signaling pathway, thereby inhibiting hepatic stellate cell activation and diminishing collagen deposition in CCl4-induced liver fibrosis models. This discovery corroborates a suggested mechanistic mechanism for the activity of telmisartan. A study published in an Egyptian pharmacology or pathology magazine affiliated with Al-Azhar University highlights telmisartan's function as an angiotensin II type 1 receptor (AT1R) antagonist with partial PPAR-γ agonistic characteristics. It emphasizes how telmisartan reduces hepatic fibrogenesis by lowering TGF-β1 levels and tissue inhibitors of metalloproteinases, while concurrently increasing matrix metalloproteinase (MMP) expression, thereby strengthening its dual antifibrotic and metabolic benefits concerning NASH-related fibrosis (Fouad, L., et al., 2022; Mousa, A. A. M., et al., 2025)

Conclusion

This study demonstrates that Telmisartan exerts a significant, dose-dependent antifibrotic effect by mitigating hepatocyte injury, reducing oxidative stress, and suppressing HSC activation. A 10 mg/kg dose proved most effective, resulting in a 65.6% reduction in fibrotic area and significant restoration of antioxidant enzymes. These findings underscore Telmisartan's potential as a dual-action therapeutic agent for the treatment of chronic fibrotic liver diseases.

References

1. Alam, S., Kabir, J., Mustafa, G., Gupta, U., Hasan, S. K., & Alam, A. K. (2016). Effect of telmisartan on histological activity and fibrosis of non-alcoholic steatohepatitis: A 1-year randomized control trial. Saudi journal of gastroenterology: official journal of the Saudi Gastroenterology Association, 22(1), 69–76. https://doi.org/10.4103/1319-3767.173762
2. Amer, F., El-rehany, M., & Othman, O. A. (2024). Combination Therapy with Vitamin D and Telmisartan to Suppress the Progression of Liver Fibrosis. Journal of Medical and Life Science, 6(2), 132-143.
3. Amoras, E. D. S. G., Monteiro Gomes, S. T., Freitas Queiroz, M. A., de Araújo, M. S. M., de Araújo, M. T. F., da Silva Conde, S. R. S., Ishak, R., & Vallinoto, A. C. R. (2020). Intrahepatic interleukin 10 expression modulates fibrinogenesis during chronic HCV infection. PloS one, 15(10), e0241199. https://doi.org/10.1371/journal.pone.0241199
4. Attia, Y. M., Elalkamy, E. F., Hammam, O. A., Mahmoud, S. S., & El-Khatib, A. S. (2013). Telmisartan, an AT1 receptor blocker and a PPAR gamma activator, alleviates liver fibrosis induced experimentally by Schistosoma mansoni infection. Parasites & vectors, 6, 199. https://doi.org/10.1186/1756-3305-6-199
5. Barr, R. G. (2014). Noninvasive liver fibrosis assessment: ElastPQ ultrasound shear wave elastography. Philips Healthcare.
6. Borém, L. M., Neto, J. F., Brandi, I. V., Lelis, D. F., & Santos, S. H. (2018). The role of the angiotensin II type I receptor blocker telmisartan in the treatment of non-alcoholic fatty liver disease: a brief review. Hypertension Research, 41(6), 394-405.
7. Bota, S., Sirli, R., Sporea, I., Focsa, M., Popescu, A., Danila, M., Strain, M., Sendroiu, M., Deleanu, A., & Dan, I. (2011). A new scoring system for prediction of fibrosis in chronic hepatitis C. Hepatitis monthly, 11(7), 548–555.
8. Chen, S. R., Chen, X. P., Lu, J. J., Wang, Y., & Wang, Y. T. (2015). Potent natural products and herbal medicines for treating liver fibrosis. Chinese medicine, 10(1), 7.
9. Fouad, L., Alzokaky, A., Abdelbaky, N., & Ahmed, K. (2022). Telmisartan inhibits the non-canonical TGF-β/JAK2/STAT3 signaling pathway and prevents carbon tetrachloride-induced liver fibrosis in rats. Azhar International Journal of Pharmaceutical and Medical Sciences, 2(1), 20-29.
10. Gan, C., Yuan, Y., Shen, H., Gao, J., Kong, X., Che, Z., ... & Xiao, J. (2025). Liver diseases: epidemiology, causes, trends and predictions. Signal Transduction and Targeted Therapy, 10(1), 33.
11. Garbuzenko, D. V. (2022). Pathophysiological mechanisms of hepatic stellate cells activation in liver fibrosis. World journal of clinical cases, 10(12), 3662.
12. Golder, M. R., Liu, J., Andersen, J. N., Shipitsin, M. V., Vohidov, F., Nguyen, H. V. T., ... & Johnson, J. A. (2018). Reduction of liver fibrosis by rationally designed macromolecular telmisartan prodrugs. Nature biomedical engineering, 2(11), 822-830.
13. Ionică, F. E., Mogoantă, L., Nicola, G. C., ChiriŢă, C., Negreş, S., Bejenaru, C., Turculeanu, A., Badea, O., Popescu, N. L., & Bejenaru, L. E. (2016). Antifibrotic action of telmisartan in experimental carbon tetrachloride-induced liver fibrosis in Wistar rats. Romanian journal of morphology and embryology = Revue roumaine de morphologie et embryologie, 57(4), 1261–1272.
14. Iwaisako, K., Jiang, C., Zhang, M., Cong, M., Moore-Morris, T. J., Park, T. J., ... & Kisseleva, T. (2014). Origin of myofibroblasts in the fibrotic liver in mice. Proceedings of the National Academy of Sciences, 111(32), E3297-E3305.
15. Kawai, M., Hongo, K., Komukai, K., Morimoto, S., Nagai, M., Seki, S., ... & Yoshimura, M. (2009). Telmisartan predominantly suppresses cardiac fibrosis, rather than hypertrophy, in renovascular hypertensive rats. Hypertension Research, 32(7), 604-610. https://doi.org/10.1038/hr.2009.61

16. Mansour, A., Eisa, M., Abdelghany, T., & Salama, S. (2022). Silymarin ameliorates diethylnitrosamine-induced liver fibrosis in Wistar rats. Al-Azhar Journal of Pharmaceutical Sciences, 66(2), 246-266.
17. Mousa, A. A. M., Helal, M. L., Sira, M. M., & Fouad, O. A. (2025). Evaluation of non-invasive serological markers of liver fibrosis in children with autoimmune hepatitis. Al-Azhar Un. Journal for Med and Virus Research and Studies. Vol 7 (3) November. 2025.
18. Yoshihara, D., Kugita, M., Sasaki, M., Horie, S., Nakanishi, K., Abe, T., ... & Nagao, S. (2013). Telmisartan ameliorates fibrocystic liver disease in an orthologous rat model of human autosomal recessive polycystic kidney disease. PLoS One, 8(12), e81480. https://doi.org/10.1038/hr.2009.61
19. Zheng, L., Zhao, Z., Lin, J., Li, H., Wu, G., Qi, X., ... & Luo, M. (2022). Telmisartan relieves liver fibrosis and portal hypertension by improving vascular remodeling and sinusoidal dysfunction. European Journal of Pharmacology, 915, 174713.