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PL-04

Evaluation of the potential protective effect of melatonin and pioglitazone on atorvastatin-induced myopathy in rats

Dalia A. Sarhan[1], Mona A. Salama[1], Rowaida R. Shehata[1], Aisha Ahmed Sherif[2], Hoda S. Hussein[1]

[1]Department of Pharmacology, [2] Department of Pathology; Medical Research Institute, Alexandria University

Introduction

Statins, intended for cardiovascular disease, are now recognized for potential anti-inflammatory and anti-cancer effects(Zhang et al., 2020).However, they can induce myopathy, often requiring treatment adjustments (Harper & Jacobson, 2010).Recent studies highlight atorvastatin-induced myopathy involving ferroptosis, a form of iron-dependent cell death, by suppressing the Nrf2-xCT/GPx4 pathway, leading to lethal lipid peroxidation (Zhang et al., 2022). ACSL4, crucial for lipid peroxidation, could be a target for therapeutic intervention (Doll et al., 2017). Melatonin and pioglitazone, known for their antioxidant and anti-inflammatory properties, have shown promise against ferroptosis-related diseases , (Andersen, 2016; Rui et al., 2021). However, their combined effect against atorvastatin-induced ferroptosis remains unexplored.

Materials and Methods

Sprague Dawley rats were treated daily with atorvastatin (Ator) (80mg/kg), melatonin (Mel) (20mg/kg), pioglitazone (Pio) (3mg/kg), Ator+Mel, Ator+Pio, and the combination of Ator+Mel+Pio for eight weeks.

Parameters of myopathy; creatine kinase (CK) and myoglobin and of mitochondrial dysfunction; lactatae dehydrogenase (LDH), were detected in the serum.

The gastrocnemius muscle was used for determination of glutathione peroxidase (GPx), malondialdehyde (MDA), citrate synthase, antioxidative transcription factor Nrf2 and Acyl-CoA synthetase long-chain family member 4 (ACSL4).

Histopathological examination was done to evaluate muscle injury by atorvastatin alone and in combination with melatonin and pioglitazone

Results

		Control (n=6)	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)	Group 4 (n=6)	Group 5 (n=6)	Group 6 (n=6)	F	p
CPK (U/L)	Mean	353.67^{d}	1154.33^{a}	363.33^{d}	379.33^{d}	899.83^{b}	913.17^{b}	781.83^{c}	172.100*	<0.001*
	± SD.	25.30	98.59	17.00	9.24	72.85	88.54	43.74		
	p_0		<0.001*	1.000	0.990	<0.001*	<0.001*	<0.001*		
	p_1			<0.001*	<0.001*	<0.001*	<0.001*	<0.001*		
	p_2				0.999	<0.001*	<0.001*	<0.001*		
	p_3					<0.001*	<0.001*	<0.001*		
	p_4						1.000	0.028*		
	p_5							0.010*		
LDH (U/L)	Mean	234.0^{d}	486.67^{a}	233.33^{d}	241.33^{d}	328.50^{bc}	357.83^{b}	301.67^{c}	99.039*	<0.001*
	± SD.	5.33	30.67	16.00	11.24	21.52	36.57	20.01		
	p_0		<0.001*	1.000	0.997	<0.001*	<0.001*	<0.001*		
	p_1			<0.001*	<0.001*	<0.001*	<0.001*	<0.001*		
	p_2				0.996	<0.001*	<0.001*	<0.001*		
	p_3					<0.001*	<0.001*	0.001*		
	p_4						0.294	0.396		
	p_5							0.002*		
Myoglobin (ng/ml)	Mean	38.33^{d}	90.0^{a}	40.25^{d}	44.02^{d}	65.97^{b}	64.48^{b}	52.57^{c}	105.771*	<0.001*
	± SD.	2.58	6.10	4.44	5.47	3.59	5.28	0.87		
	p_0		<0.001*	0.988	0.301	<0.001*	<0.001*	<0.001*		
	p_1			<0.001*	<0.001*	<0.001*	<0.001*	<0.001*		
	p_2				0.751	<0.001*	<0.001*	<0.001*		
	p_3					<0.001*	<0.001*	0.028*		
	p_4						0.997	<0.001*		
	p_5							0.001*		

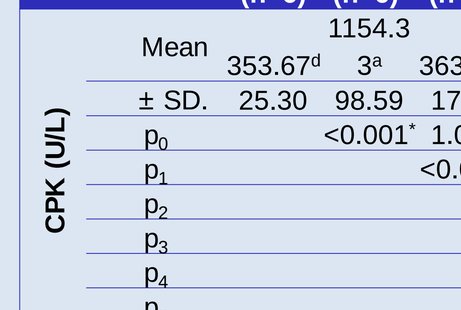

Figure (1): Comparison between the different studied groups according to GPX activity

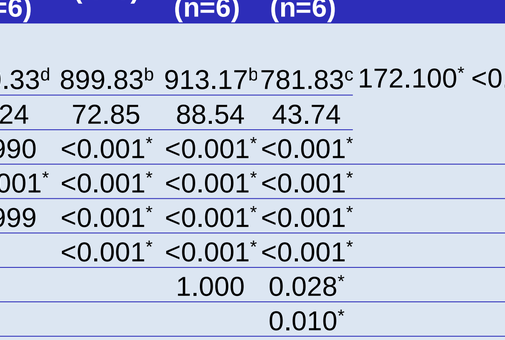

Figure (2): Comparison between the different studied groups according to MDA

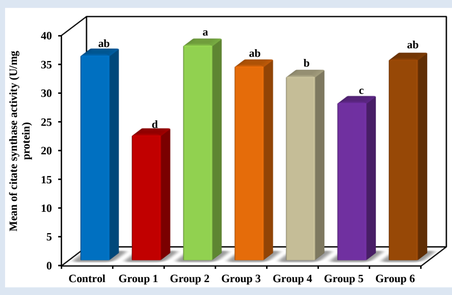

Figure (3): Comparison between the different studied groups according to Citate synthase activity

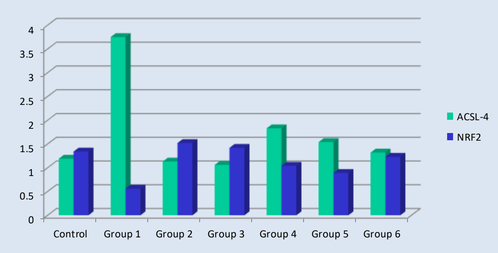

Figure (4): Effect of treatment with atorvastatin, melatonin, pioglitazone and their combination on ACSL4 and Nrf2 in gastrocnemius muscle of rats.

Conclusions

In this study, melatonin or pioglitazone with atorvastatin reduced atorvastatin- induced myopathy, mitochondrial dysfunction, and ferroptosis. Atorvastatin with melatonin enhanced Nrf2 and citrate synthase levels and reduced MDA levels better than with pioglitazone. Atorvastatin with pioglitazone significantly lowered ACSL4 levels more effectively than with melatonin, crucial in atorvastatin-induced ferroptosis.

Targeting the Nrf2/GPx/ACSL4 pathway with both reversed atorvastatin-induced muscle damage. Though not reaching normal levels, combining all three drugs improved myopathy markers significantly, highlighting the importance in statin-induced muscular injury mitigation.

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