



Investigating the Role of Angiotensin Receptor Blockers (ARBs) In High Fat Diet-Induced Sarcopenic Rats Associated Muscle Wasting

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Abstract

Sarcopenia-induced muscle loss has a significant societal impact on older people and progressively increases frailty and disability. The anabolic hormone insulin growth factor-1 (IGF-1) is a key growth hormone for muscle growth by inducing protein synthesis while catabolic muscle RING-finger protein-1 (MuRF-1) has been identified as E3 ubiquitin ligases and mediated proteolysis. The present study aimed to screen Angiotensin Receptor Blockers (ARBs) binding affinity to specified proteins through an *in-silico* approach. Out of eight ARBs, azilsartan and telmisartan exhibited good affinity to IGF-1 and MuRF-1 proteins. Then lead ARB, azilsartan (AZL) was further investigated in High-Fat Diet (HFD)-induced sarcopenia-associated muscle loss in the rat model. Male *Sprague Dawley* rats, 4 and 14 months were used as young and old, respectively, and divided into control and treatment. The control group was treated with vehicles and azilsartan treatment. Old rats were further fed with HFD for 4 months and served HFD-induced old rats. Further, HFD-induced sarcopenic rats were divided into the old control and AZL-treated old group. AZL was given at the dose of 8 mg/kg, *per oral* for 6 weeks. After treatment, rats were analyzed for functional muscle tests and results showed that AZL significantly increased muscle coordination and locomotor activities in sarcopenic rats. Next, animals were sacrificed, and gastrocnemius muscles were collected for oxidative stress and antioxidant levels. Results showed that AZL significantly restored glutathione and reduced lipid peroxidation and protein carbonyl levels. In conclusion, azilsartan treatment showed significant muscle coordination activity and restoration of antioxidants status in HFD-induced sarcopenic rats. These findings suggest that AZL could be a good intervention to prevent and treat old-age muscle wasting.

488

Keywords: Sarcopenia, azilsartan, antioxidants, High fat diet, skeletal muscle.

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Introduction

Sarcopenia is defined to have lower muscle mass and strength and decreased physical activities in the aged population (Trivedi and Khatri, 2022). According to the Asian Working Group for Sarcopenia (AWGS), the 2019 consensus diagnosing sarcopenia needs evaluation of both muscle quality and quantity (Chen et al., 2020). Further, oxidative stress, proinflammatory markers, and apoptotic cell death with obesity worsen the skeletal muscle condition in the older population (Li et al., 2022). Recently, an angiotensin receptor blocker (ARB), losartan has shown efficacy in a clinical trial for improving muscle function and

grip strength and was found to be effective in sarcopenic subjects (NCT01989793). Moreover, a new generation of the “sartan” category, azilsartan has been FDA-approved as an antihypertensive drug (Hjermitslev et al., 2017). The study reports that angiotensin II causes skeletal muscle wasting and inhibition of Angiotensin II has proven to be useful in maintaining skeletal muscle fibre composition, and endothelial function (Deminice et al., 2020, Kingsley et al., 2021, Zhou et al., 2015, Semprun-Prieto et al., 2011, Silva et al., 2019). Furthermore, perindopril, an angiotensin-converting enzyme inhibitor also showed positive results in clinical trials against the

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treatment of sarcopenia in combination with exercise (Harper et al., 2020). Overexpression of propeptides IGF-1Ea and IGF-1Eb in skeletal muscle demonstrates the effect on muscle aging and showed that these propeptide activates anabolic signaling pathway to avert the muscle loss in sarcopenia (Ascenzi et al., 2019). Further, catabolic pathways linked with MuRF-1 and was responsible for the degradation of contractile muscles proteins and might be targeting the sarcomeric proteins responsible for muscle loss in old people (Ascenzi et al., 2019, Denison et al., 2015). Additionally, computational docking is an important tool to analyze the biomolecular interactions and mechanisms on the basis of based on structure-based drug design (Forli et al., 2016). Findings demonstrate that the *in-silico* docking study provided the excellent insights to execute and analyze the molecules for skeletal muscle disease and may improve the outcomes of data (Rupert et al., 2020). Apart from this, numerous sarcopenic animal models exist such as hindlimb unloading/suspension and immobilization to study the mechanisms to explore the sarcopenia (Speacht et al., 2018). High-fat diet (HFD)-induced sarcopenia is also a validated model to study muscle loss in preclinical studies and shown to decrease the muscle strength, resistance, and loss (Abrigo et al., 2016). The study also showed that aged animals were found to decreased muscle function, altered myofibers, and leads to decreased muscle function ability in the senescence stage (Edström and Ulfhake, 2005). However, correct posture needs the—a synchronized act consisting of motor response, sensory input, cortical integration, and sciatic function index that could be helpful in assessing help access the muscle functions (Varejão et al., 2001). Moreover, oxidative stress generates the reactive oxygen species (ROS), and excessive ROS are reported to decrease the both rest and contractile activity in skeletal muscles that eventually leads to cell death (Kozakowska et al., 2015, Chen et al., 2022). These findings imply that oxidative stress is playing a pertinent role in skeletal muscle damage which may be further restored by antioxidants (Kozakowska et al., 2015). Thus, suppressing oxidative stress may be an efficient approach to preventing skeletal muscle cell death. Therefore, the present study investigates the effect of angiotensin receptor

blocker(s) against sarcopenic muscle wasting induced by high fat diet.

Materials and Methods

2.1 Chemicals: Azilsartan was obtained from a gift sample from CTX Life Sciences, Surat, India. All the chemicals were purchased from Sigma, USA, until specified. All the solvents used were of analytical grade.

2.2. *In silico* molecular docking: In the current work, molecular docking was carried out to envisage the binding affinities of an angiotensin receptor blockers (ARBs) viz. Azilsartan, Telmisartan, Losartan, Eprosartan, Irbesartan, Olmesartan, Candesartan and Valsartan as ligands with insulin growth factor-1 (IGF-1) and MuRF-1 as anabolic and catabolic proteins respectively. Firstly, the crystal structure of proteins was downloaded from the protein data bank (Berman et al., 2000). The active site of proteins was obtained from Biovia Discovery Studio Visualizer. After this, the PDBQT files for proteins and ligands were prepared and grid boxes were created using the graphical user interface programme Auto Dock Tools (ADT). ADT assigned the protein polar hydrogens, united atom Kollman charges, solvation parameters, and fragmental volumes. The prepared file was saved in PDBQT format by Auto Dock. The grid file was generated and the size was set to 60x60x60 xyz points with a grid spacing of 0.375 and a grid centre at -1.095, -1.554, and 3.894 (x, y, and z). The docking was carried out on Auto Dock/Vina was used for docking, and the binding affinities (kcal/mol) and the number of probable hydrogens and amino acids were determined. The position with the highest binding energy or affinity was selected.

2.3. Experimental design: Animal experiments were approved by the Institutional animal research committee. 11-12 months & 3-4 months old male *Sprague-Dawley (SD)* rats were procured from Central Drug Research Institute, Lucknow, India. Animals were housed according to the guidelines. Animals were fed with normal chow diet and water *ad libitum*. Animals were randomly divided into four groups (n=6). Further allocated as ±Control and Treatment. The control group includes young and high fat diet (HFD)-induced sarcopenic rats (old control) and was treated with a vehicle,



also grouped as young control (YC) and old control (OC). 14-15 months old male rats were treated with high fat diet (Bharat Ansh Scientific Industries, Lucknow) for four months. At the end of 18-19th months, rats were switched to standard chow diet (Munguia et al., 2020, Joseph et al., 2019, Bai et al., 2019). Azilsartan (8 mg/kg, oral for 6 weeks) (Alzahrani et al., 2020, Jin et al., 2014) was given as treatment to young and HFD-induced sarcopenic old rat, also grouped as young treatment (YT) and old treatment (OT). Muscle coordination tests were performed after the last day of treatment and then animals were sacrificed. Gastrocnemius muscles (GN) were collected and performed the oxidative stress and antioxidant tests.

2.3.1. Assessment of body weight and GN muscle weight

Young and old rats were weighed during the initial and termination of the experiment (Han et al., 2016). Further, GN muscle weight was also taken.

2.3.2. Assessment of muscle coordination parameters

2.3.2.1. Rotarod test: Rotarod test was used to evaluate the motor coordination. In brief, animals were acclimatized for thirty minutes for six days. Animals were placed on the rod for final evaluation, which had two plastic plates on the sides to keep them from exiting. Animals were kept for low (10 RPM), medium (20 RPM) and high (40 RPM) three times reading of falling time was computed at an interval of 15 min. Rats who remained on the rotating rod for 180 seconds received a full score (Jänicke et al., 1983).

2.3.2.2. Horizontal (hanging) wire test: This test was used to determine the muscular strength. On a 0.4 cm-diameter, 83 cm-long rod that was suspended 43 cm above a tabletop, animals were hung by their front paws. The length of hanging time (60 seconds) was calculated for each animal using two, four, or even it's tail to support itself. Three readings per animal were taken and the longest hanging duration was used for evaluation (Jänicke et al., 1983).

2.3.2.3. Actophotometer analysis: The locomotion number was evaluated by using the actophotometer test. Animals were put inside the

actophotometer and through the holes, three light beams were passed from all sides. Rats were kept in the activity cage for 5 minutes at a time, and the number of interruptions of the light beams by the rats was noted. The digital counter stopped keeping track of time when the animal stopped moving. The activity was noted when the animal began walking. The number of times the rat moved around on all fours between two opposing walls of the cage was referred to as "Walks." The amount of time spent walking was noted to analyse the results (Gosavi et al., 2020, Santhanalakshmi et al., 2021).

2.3.2.4. Footprint analysis: An impression of the rats' hind feet was stained with blue ink to assess their gait. They were then instructed to proceed through a cardboard-and-blank-paper corridor that was 100 cm long, 10 cm wide, and 20 cm high. Before the experiment began, the rats received three days of training. The trial was repeated if a rat failed to complete a full crossing. Following an air drying period in a room with good ventilation, the sheets were measured for step length and step width along each rat's entire path of travel. Step length and step width were used to express readings (cm) (Patel et al., 2012).

2.3.3. Estimation of oxidative stress (MDA, H₂O₂, PC) and antioxidant (GSH, CAT, DPPH) levels

Bradford's method was used to estimate the total protein in the rat gastrocnemius (GN) muscle. Bovine serum albumin (BSA) was used as standard (Roberson et al., 2020).

2.3.3.1. Malondialdehyde (MDA) level: Malondialdehyde was produced using trichloroacetic acid (TCA) and thiobarbituric acid (TBA) (MDA). Briefly, 0.8% thiobarbituric acid (TBA) and 30% trichloroacetic acid (TCA) was added to 100 mg of homogenized GN tissue. Following that, samples were kept in a 95°C water bath with shaking for 30 minutes. After cooling, samples were centrifuged for 15 minutes at 3000 rpm. At 540 nm, absorbance was measured in comparison to a blank, which contained no sample. Using the 1, 1, 3, 3 tetraethoxy propane, lipid peroxidation was calculated from the standard curve and expressed as nM MDA/g of protein. The amount of MDA present in a sample was calculated



according to the following equation (Ohkawa et al., 1979).

2.3.3.2. Hydrogen peroxide (H₂O₂) scavenging activity: Hydrogen peroxide scavenging activity was carried out using H₂O₂. GN samples were made up of various concentrations of distilled water, and then they were combined with 4 mM H₂O₂ solution (phosphate buffer 0.1 M, pH 7.4) and incubated for 10 min. At 230 nm, the solution's absorbance was measured in comparison to a blank solution containing a sample devoid of H₂O₂ (Oyedemi et al., 2010).

2.3.3.3. Protein carbonyl (PC) level: Protein carbonyl analysis was performed using di-nitrophenyl hydrazine (DNPH). In order to precipitate GN tissue samples, 10% TCA solution was used. The samples were then centrifuged for two minutes at 13,000 rpm to separate the supernatant. Each sample was incubated for 1-hour incubation in 0.2% diphenyl picryl hydrazine while being continuously vortexed every five minutes. Next, 100% TCA was added, vortexed, and centrifuged for 5 minutes at 13,000 rpm. Pellet was washed three times with 500 mL of an ethanol: ethyl acetate (1:1) solution after the removing supernatant. The pellet was then dissolved in 6.0 M guanidine hydrochloride, and absorbance at 360 nm light wavelength was taken (Singh et al., 2018).

2.3.3.4. Glutathione (GSH) level: The method of 5, 5'- dithiobis-2-nitrobenzoic acid (DTNB) for glutathione peroxidase (GSH) was used. 125µl of 10% tissue homogenate prepared in phosphate buffer. And add 100 µl of distilled water then added 25 µl of 50% trichloro acetic acid (TCA) and vortexed for 10 min. After vortexing, samples were centrifuged at 5000 rpm for 10min. The supernatant of samples was collected 66 µl after collection of supernatants added 131 µl Tris buffer at pH 8 and Ellman's reagents (DTNB) were added to each sample in 3 µl. Absorbance was taken at 405nm with an ELISA plate reader (Ellman, 1959, Roy et al., 2018).

2.3.3.5. Catalase activity: For estimating Catalase (CAT) activity, hydrogen peroxide was used. In the phosphate buffer (0.1M pH 7.4), a hydrogen peroxide solution was made and

incubated for 10 minutes. H₂O₂ solution was added to the sample, and the solution's absorbance was measured at 230 nm against a blank (Alam et al., 2013).

2.3.3.6. 2, 2-Diphenyl-1-picryl-hydrazyl-hydrate (DPPH) scavenging activity: DPPH was used for this activity. The DPPH-scavenging activity of the homogenate was measured according to a previously reported method (Kim et al., 2021).

3. Statistical Analysis

Statistical analysis was done with Graph Prism pad (version 8.01). For the statistical significance of the differences between the comparisons, a two-way analysis of variance (ANOVA) was performed, followed by a *post-hoc* analysis Sidak multiple comparison test. The results were presented as the mean ± standard error of mean (SEM) for each group. Statistical significance was defined as *p<0.05, **p<0.01, and ***p<0.001.

4. Results

4.1. Screening of ARBs for anabolic and catabolic proteins through *in silico* approach

All ARBs drugs *viz.* Azilsartan, Telmisartan, Losartan, Eprosartan, Irbesartan, Olmesartan, Candesartan, and Valsartan were screened by using Autodock Vina. Out of the most effective screened drugs, azilsartan & telmisartan and their binding affinity showed values of -7.2 Kcal mol⁻¹ & -7.4 Kcal mol⁻¹ respectively. The binding affinity for IGF-1 protein (10.1 Kcal mol⁻¹) & MuRF-1 (10.4 Kcal mol⁻¹ protein) were shown in Table 1 & Figure 1. Results showed that azilsartan was identified as an effective lead against IGF-1 and MuRF-1 (anabolic and catabolic respectively). For further studies, we had chosen the azilsartan as an intervention.



Table 1: Screening of all ARBs binding energies by docking for anabolic & catabolic proteins with many ARBs

S. No.	Compounds	Amino acids involved in an interaction with IGF-1	Amino acids involved in an interaction with MURF-1	Binding Affinity (Kcal mol ⁻¹) with IGF-1 (1B9G)	Binding Affinity (Kcal mol ⁻¹) with MURF-1 (3DDT)	H-Bonds with IGF-1	H-Bonds with MURF-1	Lipinski's rule of five Properties	
								Values	Values
1.	Azilsartan	GLU3, ARG42, ARG43, LYS55,	GLU8, VAL40, HIS35, LYS7	-7.2	-10.1	0	1	Molecular weight (<500 Da)	456.45 g/mol
								LogP (<5)	3.79
								H-Bond donor (≤5)	2
								H-bond acceptor (≤10)	7
								Violations	0
2.	Candesartan	GLU3, ARG43, LYS55, ARG42, ASP40, SER38	LYS30, LYS30, LYS36, CYS21, CYS29, VAL29, HIS32	-5.4	-9.3	1	1	Molecular weight (<500 Da)	440.45 g/mol
								LogP (<5)	3.53
								H-Bond donor (≤5)	2
								H-bond acceptor (≤10)	7
								Violations	0
3.	Eprosartan	PHE25, TYR24, VAL17	LYS30, GLU8, HIS35, VAL23, ALA37, PHE32	-6.5	-9.1	1	0	Molecular weight (<500 Da)	424.51 g/mol
								LogP (<5)	3.92
								H-Bond donor (≤5)	2
								H-bond acceptor (≤10)	5
								Violations	0
4.	Irbesartan	GLN15, TYR24, CYS18, PHE25, PRO28	CYS21, CYS29, HIS35, ILE34, LYS30, LYS7, VAL31, LYS30, VAL31	-6.4	-8.1	0	1	Molecular weight (<500 Da)	428.53 g/mol
								LogP (<5)	4.28
								H-Bond donor (≤5)	1
								H-bond acceptor (≤10)	5
								Violations	1
5.	Losartan	ASP40, ARG43, SER38, ARG37, LYS55, ARG42	ARG38, PRO30, CYS25, HIS37, LYS5	-6.6	-9.5	1	0	Molecular weight (<500 Da)	422.91 g/mol
								LogP (<5)	3.86
								H-Bond donor (≤5)	2
								H-bond acceptor (≤10)	5
								Violations	0
6.	Olmesartan	ARG37, SER38, ARG43, ASP40, CYS39, LEU41	LYS7, PHE30, GLU17, HIS43, GLU31	-5.5	-8.3	2	2	Molecular weight (<500 Da)	446.50 g/mol
								LogP (<5)	3.08
								H-Bond donor (≤5)	3
								H-bond acceptor (≤10)	7
								Violations	0
7.	Telmisartan	ALA8, VAL11, VAL31, PRO28, PHE25	LYS30, PHE32, GLU8, ILE34, CYS38, CYS21, ALA37, GLU38, HIS C.35	-7.4	-10.4	0	1	Molecular weight (<500 Da)	514.62 g/mol
								LogP (<5)	5.98
								H-Bond donor (≤5)	1
								H-bond acceptor (≤10)	4
								Violations	2
8.	Valsartan	ARG42, GLY1, PHE16, ALA13	VAL40, LYS30, HIS35, VAL23, LYS7, PRO24, ILE14	-4.9	-6.7	0	1	Molecular weight (<500 Da)	435.52 g/mol
								LogP (<5)	3.69
								H-Bond donor (≤5)	2
								H-bond acceptor (≤10)	6
								Violations	0



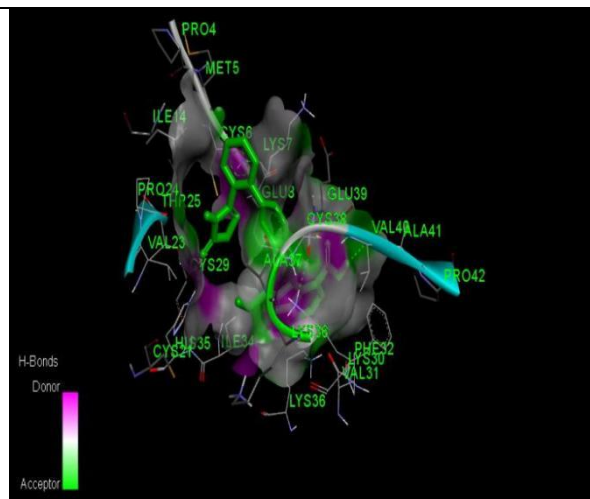
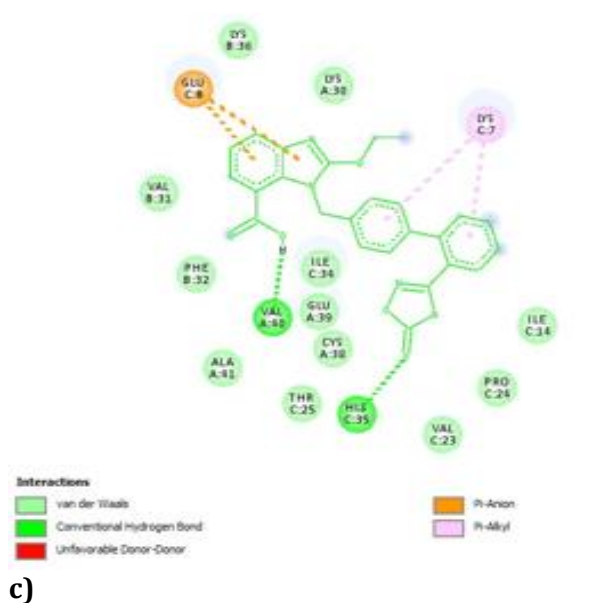
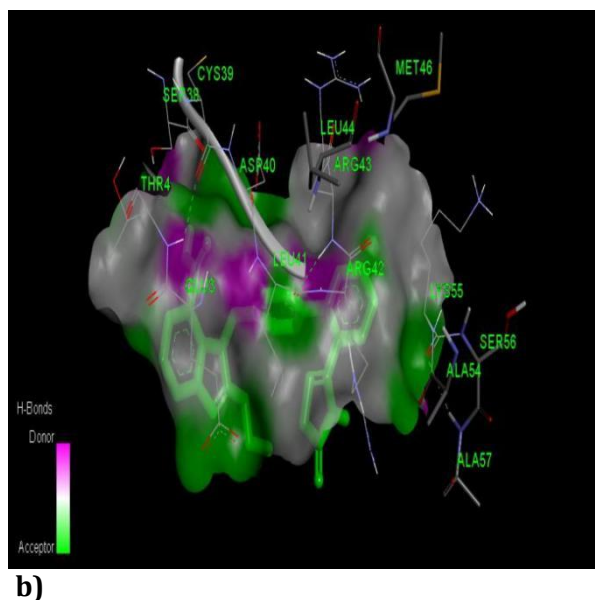
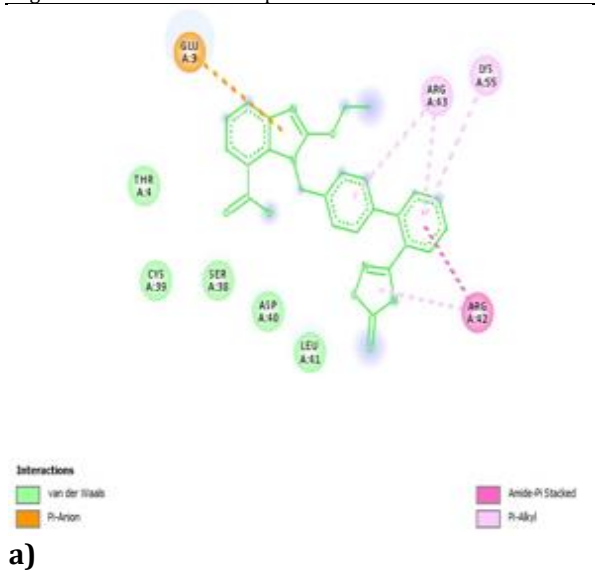
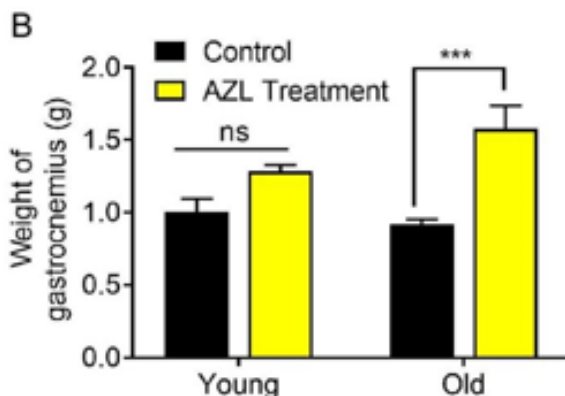
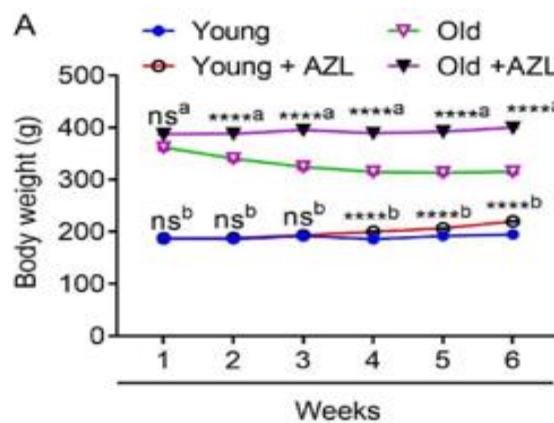


Figure 1. 2D and 3D docking images of Azilsartan for anabolic (IGF-1) and catabolic (MURF-1) proteins; a) 2 D Azilsartan for IGF-1, b) 3 D Azilsartan for IGF-1, c) 2 D Azilsartan for MURF-1, d) 3 D Azilsartan for MURF-1

4.2. Effect of azilsartan (AZL) on body weight, gastrocnemius (GN) muscle weight and GN muscle weight ratio on HFD induced sarcopenic rat

At the end of the study, young AZL & old AZL group showed significant increase in the body weight (Figure 2 A), GN muscle weight (Figure 2 B) & body weight/ GN weight ratio (Figure 2 C) as compared to age matched control group.



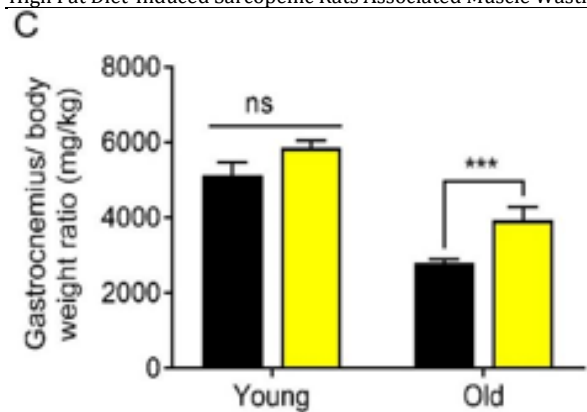


Figure 2: Effect of Azilsartan on A- body weight; B- GN muscle weight; C- GN muscle weight ratio on HFD induced sarcopenic rats

4.3. Effect of azilsartan on muscle grip strength and motor coordination in HFD-induced sarcopenic rat

Results showed that azilsartan treatment significantly improved the muscle coordination

parameters as evident in low and medium RPM (Figure 3 A). However, results were non-significant at high RPM in the young group and has significant result in old group. Next, the horizontal-wire test results were also found significant in sarcopenic AZL treated rats (OT) as compared with old control (OC) (Figure 3 B). The locomotion number in actophotometer tests of azilsartan treated sarcopenic & young rats were significantly increased when compared to respective control groups (Figure 3 C). Also, stride length was found to be improved by AZL treatment (Figure 3 D) in both right-right (R-R) and left-left (L-L) sides. Taken together, these results showed that azilsartan significantly improved the muscle coordination, muscle strength and locomotor performance in HFD-induced sarcopenic rats.

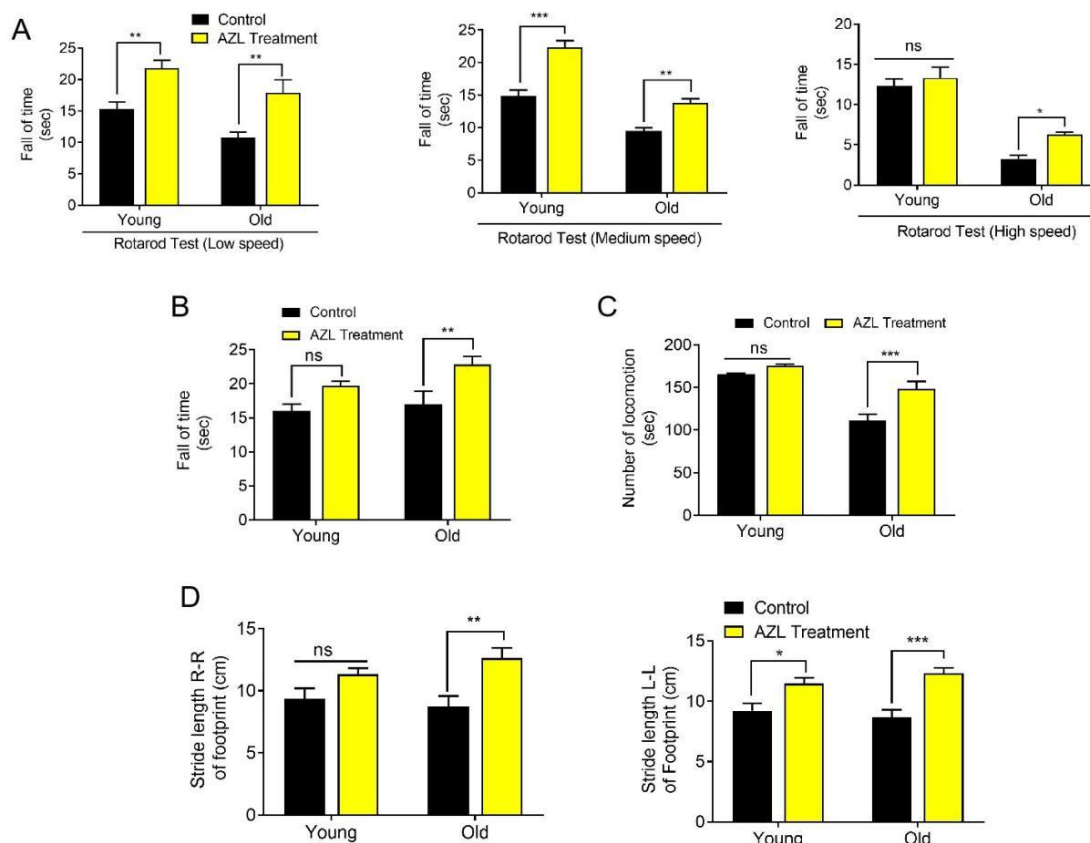


Figure 3: Effect of Azilsartan on muscle function, strength, and motor-coordination analysis on HFD induced sarcopenic rat; A- Rotarod test on Low speed, Medium and High Speeds; B- Horizontal-wire test; C- Actophotometer test; D- Footprint analysis on both Right and Left Stride lengths

4.4. Effect of azilsartan on oxidative stress and antioxidant levels in HFD-induced sarcopenic rat

Results showed that azilsartan significantly restored the GN muscle oxidative stress (MDA, PC, & H₂O₂ scavenging) (Figure 4, I; A-C) and antioxidant levels (GSH, CAT & DPPH scavenging) (Figure 4, II; D-F) in the HFD-



induced sarcopenic rat (old control rat). These results showed that AZL may act as an

antioxidant and helping to restore their level in sarcopenic muscles.

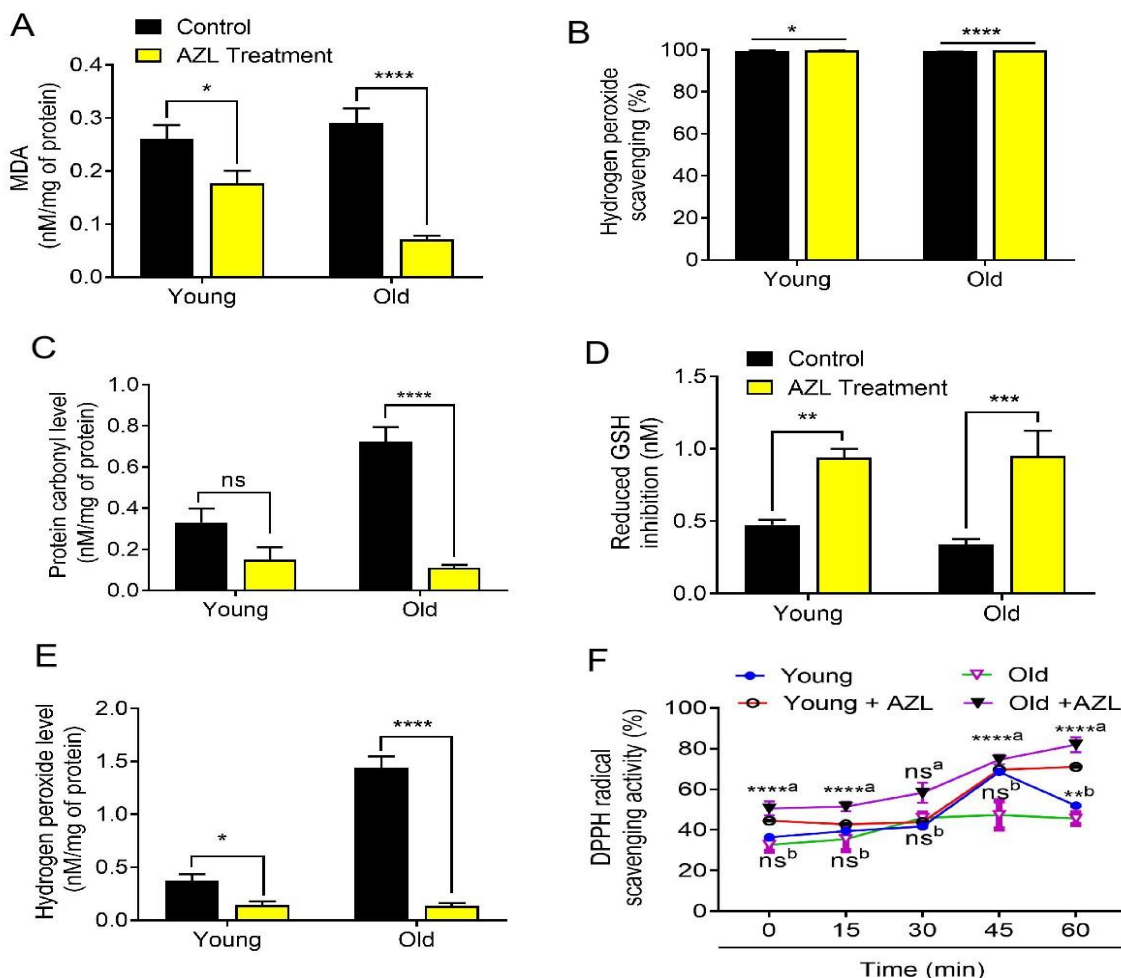


Figure 4: Effect of Azilsartan on oxidative stress and antioxidant status on HFD induced sarcopenic rat: I- Oxidative stress parameters- A- MDA level; B- H₂O₂ scavenging activity; C- Protein carbonyl level II- Antioxidants parameters- D- Reduced GSH level; E- Catalase (H₂O₂ level) activity F- DPPH scavenging activity

Discussion

As people age, sarcopenia causes a decline in physical function, which makes it harder for them to carry out daily tasks and lowers their quality of life (Yuenyongchaiwat and Akekawatchai, 2022). Therefore, the purpose of the current study is to investigate the effect of angiotensin receptor blockers (ARBs) on skeletal muscle loss in high fat diet (HFD)-induced sarcopenia. Our *in silico* study showed among all screened ARBs, both azilsartan and telmisartan were showing the good binding affinity against the insulin growth factor-1 and MuRF-1. Next, azilsartan was used for further animal studies and results showed that azilsartan significantly improved muscle performance and restored skeletal muscle loss in HFD-induced sarcopenic rats. Results

demonstrates azilsartan significantly increased the body weight, gastrocnemius muscle weight, and motor coordination function. Further, azilsartan treatment notably restored the levels of antioxidants and oxidative stress in skeletal muscles, which showed the effectiveness of azilsartan against old age associated muscle wasting. The study found that diet plays an important role in weight loss (muscle, fat, and bone mass) and that dietary changes exacerbate age-related sarcopenia and frailty outcomes in older people (Colleluori and Villareal, 2021). Additionally, as people age, their strength declines much more quickly than their muscle mass does, indicating a decline in the quality of their muscles (Hiol et al., 2021). The two most important physical function indicators for sarcopenia detection are skeletal muscle mass



and gait speed (Kwon et al., 2019). Accordingly, our findings demonstrated that azilsartan increased the grip strength, number of locomotions and fall of time when compared to age matched control. Furthermore, azilsartan also increased stride length on both the left and right sides of HFD-induced old rats. This finding may suggest that azilsartan improves integrated physical performances in sarcopenic rats by increasing gait speed and improving movement coordination. Azilsartan, in particular, may aid in preventing the onset and progression of HFD-induced sarcopenia.

Reactive oxygen and nitrogen species (ROS/RNS), also known as oxidative stress, can accumulate over time and lead to age-related declines in skeletal muscle quantity and quality. Accumulation of ROS/RNS can cause redox modifications to nucleic acids, lipids, and proteins, resulting in macromolecular damage, proteolysis and/or mitochondrial dysfunction (Di Meo et al., 2016). However, until recently, the mechanistic relationship between age-induced oxidative stress and muscle strength loss was unknown. A study demonstrated how oxidative stress can lower muscle quality by impairing activation at the neuromuscular junction and muscle quantity by shifting protein balance into a deficit (Kozakowska et al., 2015). Studies on ageing rats found that supplementing with antioxidants could prevent the loss of muscle mass, including sarcoplasmic and myofibrillar proteins (Cholewa et al., 2017). Furthermore, ROS production can accelerate the breakdown of proteins by activating muscle proteases allosterically or by upregulating ubiquitin-proteasome system components (i.e., caspases and calpains) (Gomez-Cabrera et al., 2020). Increased expression and content of the proteasome are linked to ageing and extended inactivity. Contrarily, oxidative stress refers to a state in which the antioxidant system is unable to adequately reduce reactive oxygen or nitrogen species due to an excess production of pro-oxidant molecules by the cell (Jordan et al., 2021, Bashan et al., 2009). Studies have shown that oxidative stress is a significant factor in skeletal muscle exercise, and that it may harm the muscle (Jamurtas, 2018). Additionally, our findings indicated that azilsartan enhanced muscle antioxidant status, indicating that an imbalance between pro- and anti-oxidants is a major contributor to the emergence of several

age-related pathological conditions, including muscle atrophy (Ziada et al., 2020, Singh et al., 2022). In addition to their potential involvement in biochemical processes, the beneficial effects may be primarily due to the intrinsic antioxidant activity of ARBs. Reactive oxygen species (ROS) production and lipid peroxidation have been shown to be significantly reduced by azilsartan (Liu et al., 2016). It is also possible that increased reactive oxygen species production may cause sarcopenia by preventing the synthesis of muscle proteins. Moreover, more studies are required to decipher the role and mechanism of oxidative stress as a determinant factor in muscle loss in ageing or not. Besides, azilsartan treatment to sarcopenic rats receiving HFD treatment restored the oxidative stress implying that azilsartan may prevent skeletal muscle functional abnormalities and their associated oxidative damage.

Conclusions

Our findings imply that treatment with azilsartan may slow the onset and progression of sarcopenia by improving muscle strength, weight and functionality in HFD induced sarcopenic rats. Notably, azilsartan restores equilibrium between oxidative and anti-oxidative levels in sarcopenic, older muscles. Hence, our findings imply that azilsartan may uncover the potential targets for interventions in the study of age-associated muscle loss.

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