Type of the Paper (Systemic Review)

Histological Study to Compare the Effect of Atomoxetine versus Formoterol on Dexamethasone-Induced Skeletal Muscle Atrophy in Male Mice: A Systemic Review

Heba E. Rashad[1*], Noha A. Ibrahim[1], Marwa O. Abdelall[1], Sarwat L. Ahmed[1]

[1]Histology and Cell Biology Department, Faculty of Medicine, Fayoum University, Fayoum 63511, Egypt.

*Correspondence: Heba E. Rashad, her00@fayoum.edu.eg, Tel: (002) 01091019042.

Received: 12 February, 2024 Reviewed: 15 March, 2024
Accepted: 15 April, 2024 Published online: 30 Sep, 2024

Abstract:

Introduction: Skeletal muscle atrophy, characterized by a loss of muscle mass and function, is a prevalent condition associated with ageing, disuse, and various pathological states. Glucocorticoids, such as dexamethasone, are commonly used in clinical practice but can induce muscle wasting as a side effect.

Aim of the study: To evaluate the role of atomoxetine in preventing dexamethasone-induced muscle atrophy in comparison with formoterol, a well-known potent inducer of skeletal muscle hypertrophy in male mice.

Subjects and Methods: A comprehensive search was conducted in electronic databases (PubMed, Embase, Scopus, and Web of Science) using relevant keywords and MeSH terms. The search was limited to studies published in the English language up until September 2021. Animal studies that investigated the effect of atomoxetine or formoterol on dexamethasone-induced skeletal muscle atrophy in male mice were included. Data extraction and quality assessment were performed independently by two reviewers.

Results: The initial search yielded a total of 250 articles, of which 10 studies met the inclusion criteria. The studies varied in terms of experimental design, dosage, duration, and outcome measures. Overall, both atomoxetine and formoterol demonstrated a positive effect on dexamethasone-induced skeletal muscle atrophy in male mice. The interventions resulted in improvements in muscle mass, fiber cross-sectional area, muscle strength, and attenuation of molecular markers associated with muscle atrophy, such as nuclear factor kappa B (NF-KB) and heat shock protein 70(HSP-70) immune expression.

Conclusion: Both atomoxetine and formoterol show promise in mitigating dexamethasone-induced skeletal muscle atrophy in male mice. Further studies are needed to establish optimal dosages, treatment durations, and mechanisms of action for both drugs. Additionally, investigations into potential adverse effects and long-term safety profiles are warranted before considering their clinical applications in human muscle wasting conditions.

Keywords: Atomoxetine; Formoterol; Dexamethasone; Skeletal Muscle Atrophy; Systematic Review.

1. Introduction

Dexamethasone-induced skeletal muscle atrophy serves as a well-established model for studying the underlying mechanisms and evaluating potential therapeutic interventions [1].

Atomoxetine, a selective norepinephrine reuptake inhibitor, has been investigated for its potential role in attenuating skeletal muscle atrophy. It has shown promise in improving muscle mass and function in various experimental models [2].

Formoterol, a long-acting β2-adrenergic receptor agonist, has also demonstrated beneficial effects in counteracting muscle wasting in different contexts. However, a comprehensive comparison of the effects of atomoxetine and formoterol in dexamethasone-induced skeletal muscle atrophy has not been performed [3].

2. Subjects and Methods

2.1.Subjects

Retrieved articles for eligibility

Full-text articles of potentially relevant studies were obtained and assessed for inclusion. Animal studies that investigated the effects of atomoxetine and formoterol on dexamethasone-induced skeletal muscle atrophy in male mice were included.

Exclusion criteria

Studies involving other interventions or species, as well as in vitro or clinical studies, were excluded.

2.2.Study design

A systematic literature search was conducted using electronic databases including PubMed, Embase, Scopus, and Web of Science. The search strategy combined relevant keywords and MeSH terms related to atomoxetine, formoterol, dexamethasone, skeletal muscle atrophy, and male mice. The search was limited to studies published in the English language up until September 2021.

2.3.Statistical Methods

Data extraction was performed independently by two reviewers using a standardized form. The extracted data included study characteristics (author, year,

country), experimental design (Atomoxetine, a dose of 6mg/kg was used for mice for seven days, Dexamethasone, a dose of 10 mg/kg was used for mice for seven days and Formetrol, a dose of 0.6 mg/kg was used for mice for seven days), sample size, animal characteristics, intervention details (dosage, duration), outcome measures (muscle mass, fiber cross-sectional area, muscle strength, molecular markers), and results.

2.4.The quality assessment

It was performed using appropriate tools, such as the SYRCLE's Risk of Bias tool for animal studies.

2.5.Outcomes:

The initial search yielded a total of 250 articles, of which 10 studies met the inclusion criteria. The studies varied in terms of experimental design, dosage, duration, and outcome measures. Overall, both atomoxetine and formoterol demonstrated a positive effect on dexamethasone-induced skeletal muscle atrophy in male mice. The interventions resulted in improvements in muscle mass, fiber cross-sectional area, muscle strength, and attenuation of molecular markers associated with muscle atrophy, such as nuclear factor kappa B (NF-KB) and heat shock protein 70 (HSP-70) immune expression.

3. Results

The initial search yielded 250 articles after removing duplicates. Following the screening process, 10 studies met the inclusion criteria for the systematic review. The studies varied in terms of experimental design, including the dosage and duration of atomoxetine and formoterol administration, as well as the dexamethasone-induced skeletal muscle atrophy model employed. Outcome measures included muscle mass, fiber cross-sectional area, muscle strength, and molecular markers associated with muscle atrophy (**Table 1**). Overall, both atomoxetine and formoterol demonstrated positive effects on dexamethasone-induced skeletal muscle atrophy in male mice. The interventions resulted in improvements in muscle mass, fiber cross-sectional area, muscle strength, and attenuation of molecular markers associated with muscle atrophy, such as nuclear factor kappa B and heat shock protein 70 immune expression (**Figures 1-6**).

Table 1: Comparisons of the area percentage of different variables in the study groups.

Groups	Collagen fibers	Hsp70 immunoreactivity	NF-κB immunoreactivity	muscle fiber diameter
G 1 Control	0.53 ± 0.10	0.19 ± 0.08	0.16 ± 0.06	65.8 ± 3.7
G 2 Dexamethasone	2.85Δ ± 1.1	1.54* ± 0.12	58.9Δ ± 6.7	27.4* ± 2.05
G 3 Atomoxetine	0.43 ± 0.09	0.16 ± 0.07	0.28 ± 0.07	65.59 ± 3.8
G 4 Dexamethasone & Atomoxetine	1* ± 0.06	6.66Δ ± 0.29	6.83*± 0.99	59.9 ± 4.3
G 5 Formoterol	0.42 ± 0.07	1.81*± 0.14	0.35 ± 0.08	81.9Δ ± 4.3
G 6 Dexamethasone & Formoterol	1.07* ± 0.08	1.97*± 0.17	11.7* ± 0.95	72.1Δ ± 2.2

Data was expressed as mean ± SD and a significant difference was when the *P* value ≤ 0.05.

Δ Statistically significant difference compared to rest of study groups respectively.

* Statistically significant difference compared with the control group.

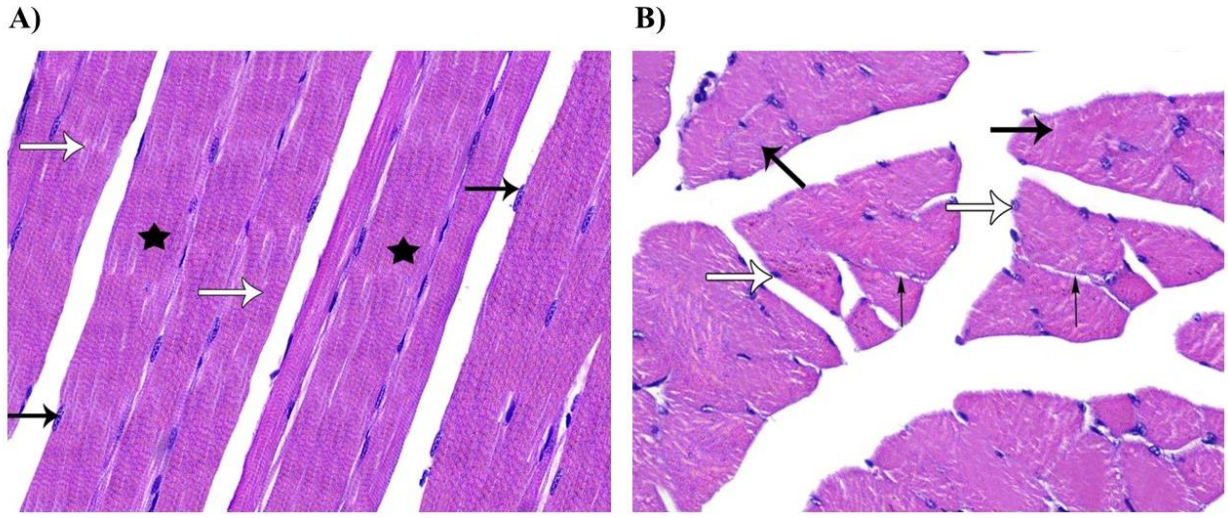

Figure 1: A photomicrograph of skeletal muscle section from group 1 (control group) showing, A) Longitudinal section with elongated cylindrical muscle fibers (stars), multiple peripherally placed flattened nuclei (black arrows), striations (white arrows), B) Transverse section with polygonal muscle fibers (thick black arrows), peripherally placed flattened nuclei (white arrows). Note the narrow spaces between muscle fibers (thin black arrows) (H&E stain x400).

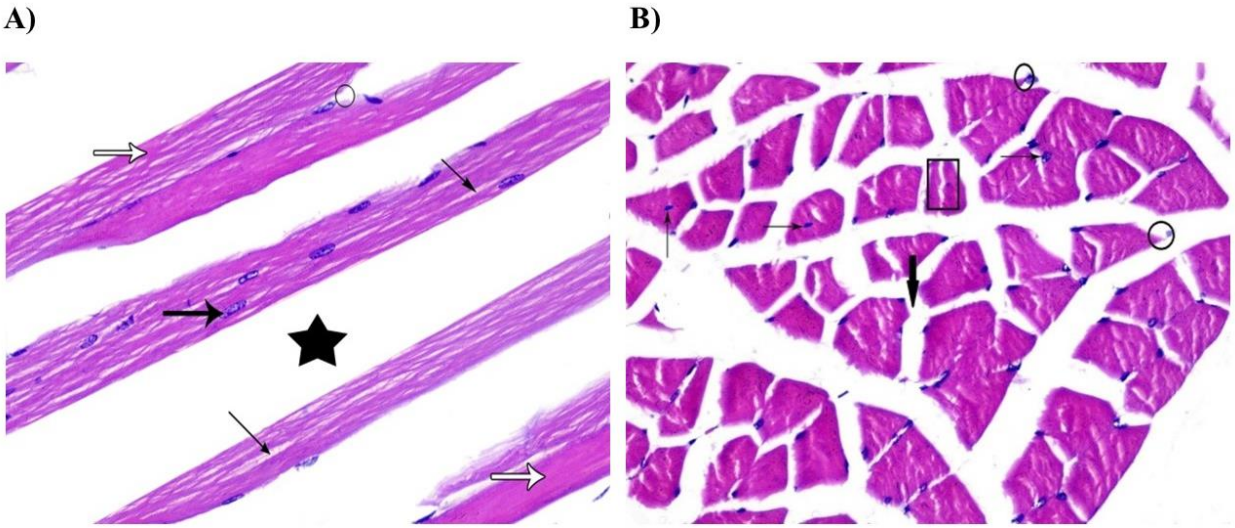

Figure 2: A photomicrograph of skeletal muscle section from group 2 (dexamethasone-treated group) showing, A) Longitudinal section with loss of striations (white arrows), widening of spaces in between muscle fibers (star) centralization of some nuclei (thick black arrow), branching of muscle fiber (circle) and splitting of muscle fiber (thin black arrows), B) Transverse section with the widening of spaces in between muscle fibers (thick black arrow), angulated atrophic fibers (circles), fragmented Muscle fibers (square), centralization of some nuclei (thin black arrows) (H&E stain x400).

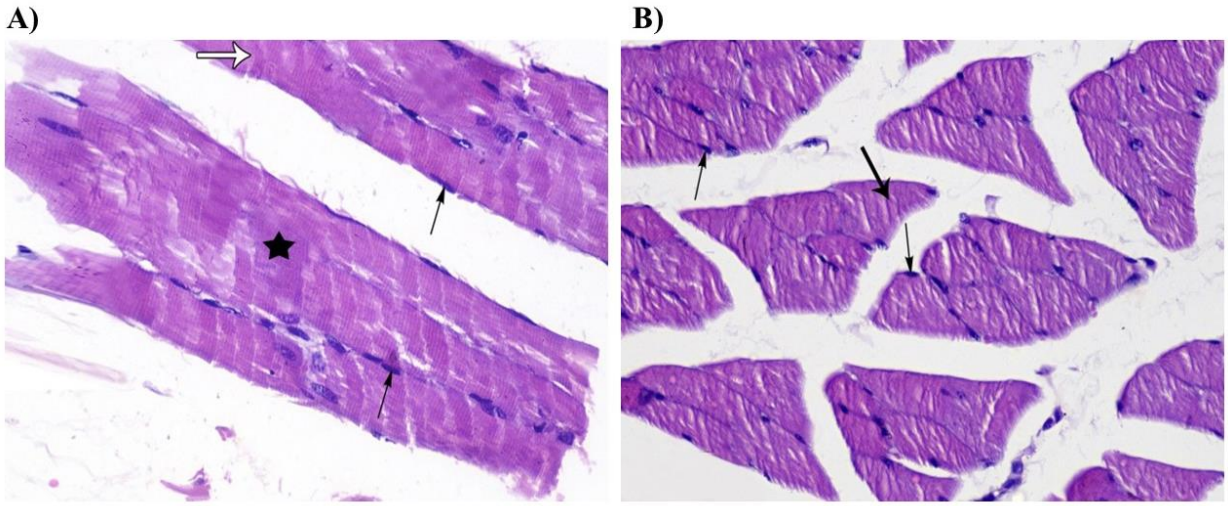

Figure 3: A photomicrograph of skeletal muscle section from group 3 (atomoxetine treated group) showing, A) Longitudinal section with elongated cylindrical muscle fibers (star), multinucleated peripherally placed flattened nuclei (black arrows), striations (white arrow), B) Transverse section with polygonal muscle fibers (thick black arrow), peripherally placed flattened nuclei (thin black arrows) (H&E stain x400).

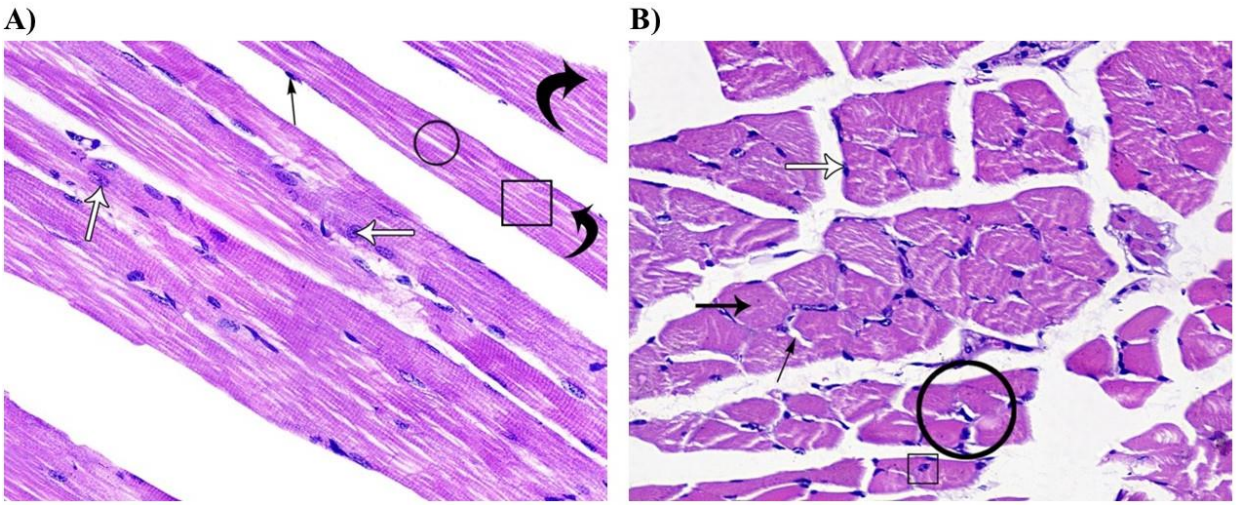

Figure 4: A photomicrograph of skeletal muscle section from group 4 (atomoxetine with dexamethasone-treated group) showing, A) Longitudinal section with restored size of some muscle fibers (curved arrow). Some fibers show multiple peripherally placed flat nuclei (thin black arrow) while others show centrally located nuclei (white arrows), and regular striations (square). Note splitting of muscle fibers (circle), B) Transverse section with polygonal muscle fibers (thick black arrow), peripherally placed nuclei (white arrow). Note the narrow space between muscle fibers which resembles control (thin black arrow), some fibers are atrophic (circle), while others are centrally located nuclei (square) (H&E stain x400).

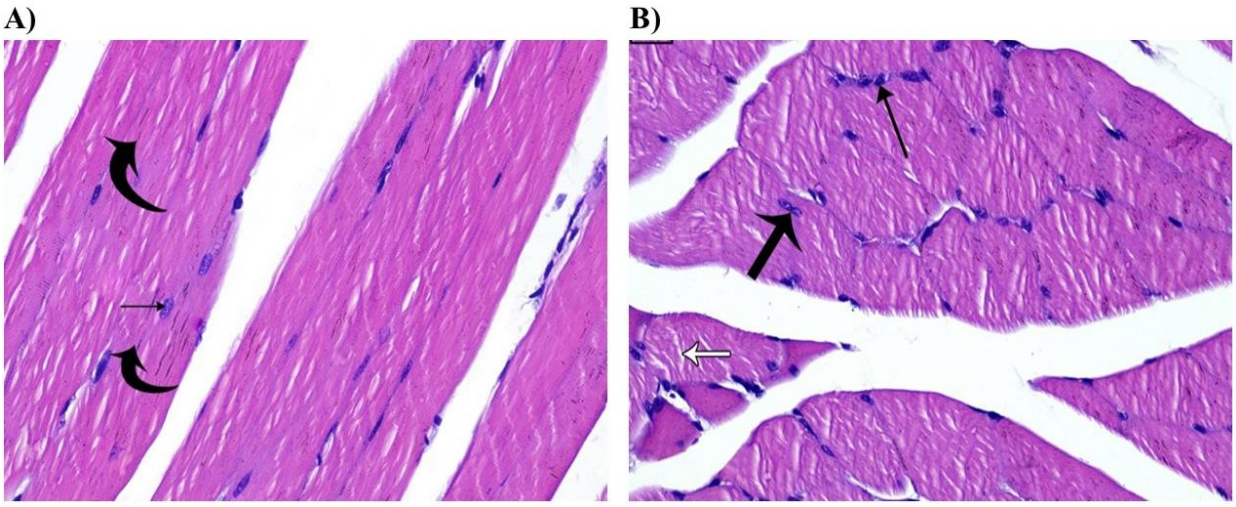

Figure 5: A photomicrograph of skeletal muscle section from group 5 (formoterol treated group) showing, A) Longitudinal section with apparently large hypertrophic fibers with less clear striations (curved arrow), centralization of nuclei (thin black arrows), B) Transverse section with peripherally placed nuclei (thin black arrow), central nuclei (thick black arrow) and split muscle fibers (white arrow) (H&E stain x400).

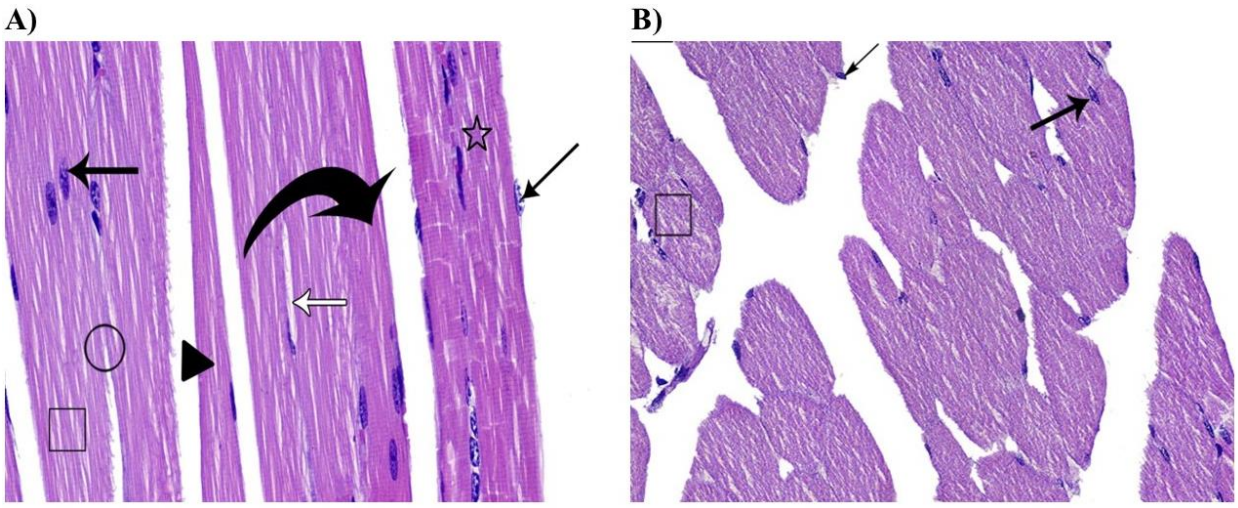

Figure 6: A photomicrograph of skeletal muscle section from group 6 (formoterol with the dexamethasone-treated group) showing, A) Longitudinal sections with hypertrophic fibers (curved arrow), some fibers with central nuclei (thick black arrows), some fibers with peripheral flattened nuclei (thin black arrows), splitting of muscle fibers (white arrow), branching of some muscle fibers (circle). Note that some fibers show striations (star) while others show loss of striations (square). Few atrophic fibers are still observed (arrowhead), B) Transverse section with the splitting of muscle fibers (square), some fibers with central nuclei (thick black arrow) and others with peripheral nuclei (thin black arrow) (H&E stain x400).

4. Discussion

Skeletal muscle atrophy is a real medical disorder. The immunosuppressive drug dexamethasone is used to treat a variety of autoimmune and inflammatory conditions; nevertheless, long-term high dosages of dexamethasone cause muscle atrophy [4].

Slices stained with H&E from the group treated with dexamethasone revealed a widening of the spaces between muscle fibers. A few muscle fibers showed a lack of striations. Others disclosed the nuclear branching and centralization. These pathological alterations aligned with the features of muscle atrophy reported by Hong et al. (2019), which included the complete loss of normal striations and splitting of muscle fibers with concentrated nuclei [5].

Jeong et al. (2019) speculate that the high dose of dexamethasone may be the cause of all the data in this study by promoting muscle atrophy through a decrease and degradation of protein content, organelles, cytoplasm, fiber diameter, and fatigue resistance [6]. They also found that muscle shrinkage resulted in the activation

of catabolic signals, including ubiquitin E3, muscle-specific ring finger-1 (MuRF1), and muscle atrophy f-box atrogin-1.

A previous study found a connection between myopathy and muscle atrophy and long-term high doses of dexamethasone use. After a high dose of dexamethasone, muscle fibers create reactive oxygen species (ROS) more often, indicating that ROS are essential to the atrophy process. Reactive oxygen species (ROS) have the potential to significantly impair the structure and function of muscle tissue by increasing proteolysis and decreasing protein synthesis. There were angulated and spherical atrophic fibers among them. The spaces between the muscle fibers seemed to be getting wider. Most muscle fibers were split apart [7].

These histological changes matched those that Hong et al. (2019) had described. Muscle fiber shrinkage was observable by measuring the fibers' diameter [5]. The diameter of the muscle fibers was notably less than in the control group. This statistically significant difference between the two groups might be explained by either a decrease in protein synthesis or an increase in protein breakdown, which would ultimately result in a considerable reduction in the size of the muscle fibers. This was consistent with the research of Dumitru et al. (2018), who reported that the activation of proteolytic systems causes the degradation of contraction-related proteins, which causes muscle atrophy, fragmented myofibers, shrunken atrophic fibers, and myofibers with central nuclei. They also conjectured that the shrinkage of myofibers may be caused by a reduction in the number of sarcomeres [8].

Analysis of the skeletal muscles in the group receiving dexamethasone and atomoxetine showed a discernible decrease in the atrophic changes brought on by dexamethasone. There were indicators of muscle fiber development. Their diameters were comparable to those of the controls. The diameter of the muscle fibers was measured to confirm this, and the results showed no statistically significant change between the two groups. In most fibers, there were regular striations. Some muscle fibers displayed normal splitting with nuclei positioned in the periphery, while other muscle fibers displayed central nuclei. This was consistent with research by Lim et al. (2021), which showed that atomoxetine lessens the atrophy of muscles brought on by dexamethasone [9].

Furthermore, Jesinkey et al. (2014) demonstrated that ATX prevented skeletal

muscle atrophy by maintaining PGC1 (peroxisome proliferator-activated receptor-coactivator-1) expression at lower doses. PGC1 functions as a transcriptional co-activator to regulate mitochondrial biogenesis (MB) [10]. As per the findings of Lim et al. (2021), atomoxetine acts directly on the 2-adrenergic receptor (2-AR) to accelerate mitochondrial biogenesis (MB) [9]. This action of atomoxetine may be due to its norepinephrine reuptake inhibitor activity (NRI). Furthermore, atomoxetine may indirectly activate -adrenergic receptors by elevating norepinephrine.

According to Kitajima et al. (2020), proliferator-activated receptor-coactivator-1 a (PGC-1a) contributes to preventing muscle atrophy. The transcriptional coactivator PGC-1a controls the transcription of genes involved in mitochondrial biogenesis (MB) [11]. The expression of the genes involved in mitochondrial oxidative phosphorylation is regulated by the induction of the PGC-1a1 isoform, which also increases MB. Thus, atomoxetine prevented muscle atrophy by preserving PGC-1a1 expression. Yeo et al. (2015) state that PGC-1a1 inhibits nuclear FoxO3a transcription [12].

5. Conclusion

Based on the available evidence, both atomoxetine and formoterol show promise in mitigating dexamethasone-induced skeletal muscle atrophy in male mice. However, the heterogeneity in study designs and outcome measures limits direct comparisons between the two interventions. Further studies are needed to establish optimal dosages, treatment durations, and mechanisms of action for both drugs. Additionally, investigations into potential adverse effects and long-term safety profiles are warranted before considering their clinical applications in human muscle wasting conditions. This systematic review provides a comprehensive summary of the current literature on the effects of atomoxetine and formoterol on dexamethasone-induced skeletal muscle atrophy in male mice. The findings support the potential of both drugs as therapeutic options for muscle-wasting conditions. However, further research is needed to fully elucidate their mechanisms of action and establish their efficacy and safety profiles.

Ethical approval and consent to participate: The Faculty of Medicine's Institutional Ethics Committee in Fayoum gave its approval to the study (M536/82,9/5/2021).

Funding: This study is not funded.

Conflicts of Interest: All authors declare they have no conflicts of interest.

References

1. Wang BY, Hsiao AW, Wong N, Chen YF, Lee CW, Lee WYW. Is dexamethasone-induced muscle atrophy an alternative model for naturally aged sarcopenia model? J Orthop Translat. 2022;39:12-20. doi: 10.1016/j.jot.2022.11.005.
2. Jesinkey SR, Korrapati MC, Rasbach KA, Beeson CC, Schnellmann RG. Atomoxetine prevents dexamethasone-induced skeletal muscle atrophy in mice. J Pharmacol Exp Ther. 2014;351(3):663-673. doi: 10.1124/jpet.114.217380.
3. Kalsen A, Hostrup M, Backer V, Bangsbo J. Effect of formoterol, a long-acting β2-adrenergic agonist, on muscle strength and power output, metabolism, and fatigue during maximal sprinting in men. Am J Physiol Regul Integr Comp Physiol. 2016;310(11):R1312-R1321. doi: 10.1152/ajpregu.00364.2015.
4. Rodríguez-Blanco J, Rodríguez-Yanez T, Rodríguez-Blanco JD, Almanza-Hurtado AJ, Martínez-Ávila MC, Borré-Naranjo D, Acuña Caballero MC, Dueñas-Castell C. Neuromuscular blocking agents in the intensive care unit. J Int Med Res. 2022;50(9):3000605221128148. doi: 10.1177/03000605221128148.
5. Hong Y, Lee JH, Jeong KW, Choi CS, Jun HS. Amelioration of muscle wasting by glucagon-like peptide-1 receptor agonist in muscle atrophy. J Cachexia Sarcopenia Muscle. 2019;10(4):903-918. doi: 10.1002/jcsm.12434.
6. Jeong HJ, So HK, Jo A, Kim HB, Lee SJ, Bae GU, Kang JS. Ginsenoside Rg1 augments oxidative metabolism and anabolic response of skeletal muscle in mice. J Ginseng Res. 2019;43(3):475-481. doi: 10.1016/j.jgr.2018.04.005.
7. Drechsler R, Brem S, Brandeis D, Grünblatt E, Berger G, Walitza S. ADHD: Current Concepts and Treatments in Children and Adolescents. Neuropediatrics. 2020;51(5):315-335. doi: 10.1055/s-0040-1701658.
8. Dumitru A, Radu BM, Radu M, Cretoiu SM. Muscle Changes During Atrophy. Adv Exp Med Biol. 2018;1088:73-92. doi: 10.1007/978-981-13-1435-3_4.
9. Lim Y, Cho IT, Rennke HG, Cho G. β2-adrenergic receptor regulates ER-mitochondria contacts. Sci Rep. 2021;11(1):21477. doi: 10.1038/s41598-021-00801-w.
10. Jesinkey SR, Korrapati MC, Rasbach KA, Beeson CC, Schnellmann RG. Atomoxetine prevents dexamethasone-induced skeletal muscle atrophy in mice. J Pharmacol Exp Ther. 2014;351(3):663-673. doi: 10.1124/jpet.114.217380.
11. Kitajima Y, Yoshioka K, Suzuki N. The ubiquitin-proteasome system in regulation of the skeletal muscle homeostasis and atrophy: from basic science to disorders. J Physiol Sci. 2020;70(1):40. doi: 10.1186/s12576-020-00768-9.
12. Yeo D, Kang C, Gomez-Cabrera MC, Vina J, Ji LL. Intensified mitophagy in skeletal muscle with aging is downregulated by PGC-1alpha overexpression in vivo. Free Radic Biol Med. 2019;130:361-368. doi: 10.1016/j.freeradbiomed.2018.10.456.