New Approaches in Exercise Physiology (NAEP)
Vol. 6, No.11, June 2024
www.nass.atu.ac.ir
DOI: 10.22054/nass.2025.85287.1174

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Exercise Reversing Aging Process and Inducing Longevity by FOXO Proteins Family

Ashrafsadat Anvarimanshadi – Faculty of Physical Education and Sport Sciences, Exercise Physiology, Allameh Tabataba'i University

Behzad Hajizadeh Maleki Department of Exercise Physiology and Sports Therapy, Kugelberg 62, Justus-Liebig-University, Giessen, Germany.

Nasrin Rafizadeh Faculty of Physical Education and Sports Sciences, Allameh Tabataba'i University, Tehran, Iran.

Davood Sabour Faculty of Physical Education and Sports Sciences, Allameh Tabataba'i University, Tehran, Iran.

Elnaz Safikhani Faculty of Physical Education and Sports Sciences, Allameh Tabataba'i University, Tehran, Iran.

Amirhossein Jafari Faculty of Physical Education and Sports Sciences, Allameh Tabataba'i University, Tehran, Iran.

– Corresponding Author: asanvary@gmail.com

How to Cite: Anvarimanshadi, A., Hajizadeh Maleki, B., Rafizadeh, N., Sabour, D., Safikhani, E., & Jafari, A. (2024). Exercise Reversing Aging Process and Inducing Longevity by FOXO Proteins Family, *Journal of New Approaches in Exercise Physiology*, 6(11), 151-180.
DOI: 10.22054/nass.2025.85287.1174

Review article
Received: April 17, 2025 Accepted: May 17, 2025

Abstract

Aging is influenced by a wide range of biological processes, with increasing attention focused on physical exercise as an effective, non-pharmacological intervention to support healthy aging and extend lifespan. This review examines how exercise influences aging mechanisms by activating Forkhead Box O (FOXO) proteins—especially FOXO3, a critical regulator of oxidative stress resistance, metabolism, and cellular repair. By integrating evidence from both animal models and human studies, we explore how different types of physical activity, including endurance and resistance training, enhance FOXO signaling pathways that govern mitochondrial health, autophagy, and inflammation control. FOXO activation appears to play a central role in mitigating age-related diseases such as sarcopenia, cardiovascular disease, neurodegeneration, and cancer. While preclinical studies consistently demonstrate these protective effects, human data are more variable, highlighting the influence of age, sex, and intervention type. Understanding the molecular connection between exercise and FOXO activity can help develop targeted exercise recommendations to promote longevity and delay age-associated functional decline.

Keywords: FOXO Proteins, Longevity, Exercise, Aging, Metabolic Health.

Introduction

Aging is a complex and multifaceted process involving numerous interconnected molecular pathways, among which the Forkhead Box O (FOXO) transcription factors play significant roles. FOXO proteins regulate crucial aspects of cellular function, including stress resistance, metabolic health, and longevity (Guan et al., 2025; Santos et al., 2023) . Physical exercise has emerged as an effective strategy to activate these proteins, providing significant anti-aging benefits through mechanisms involving mitochondrial enhancement, metabolic regulation, and improved stress responses (Williamson et al., 2010; Zeng et al., 2020). Despite the promising evidence linking exercise and FOXO activation to improved aging outcomes, critical gaps in knowledge remain. Translating findings from animal models to human populations is challenging, particularly regarding the optimal types, intensities, and durations of exercise necessary to stimulate FOXO pathways effectively (Fan et al., 2017; Jin et al., 2023). Additionally, existing research highlights variations in FOXO responsiveness due to factors such as age, genetic background, and individual health conditions (Silva-Sena et al., 2018; Williamson et al., 2010). Addressing these controversies and understanding their implications are vital for developing targeted exercise guidelines tailored specifically to older adults, with the ultimate goal of extending healthy lifespan and mitigating age-related diseases.

Methods

This systematic review aimed to explore how various forms of physical exercise influence the aging process via activation of FOXO proteins, particularly FOXO3. The review followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (see Figure 1).

Rationale for Including Animal and Human Studies

Given the molecular focus of this review, both animal and human studies were included to bridge mechanistic findings with potential

clinical relevance. Animal models offer detailed insight into cellular and molecular pathways, while human studies provide valuable evidence of physiological outcomes. Integrating both allows for a more comprehensive understanding of FOXO-mediated exercise effects across species and contexts.

Eligibility Criteria

- inclusion and exclusion criteria were defined using the PICOS framework (Table 1).
- Population: Studies on animals or humans investigating aging and FOXO protein activity.
- Intervention: Exercise in any form (aerobic, resistance, endurance, etc.).
- Control: Sedentary or baseline comparison groups.
- Outcomes: Changes in FOXO protein expression or activity, especially FOXO3.

Only original research articles were included. Reviews, commentaries, and letters were excluded.

Information Sources and Search Strategy

A systematic search was conducted using PubMed, Scopus, and Web of Science databases. Keywords included combinations of "FOXO proteins," "exercise," "aging", "longevity", "FOXO3", "oxidative stress", and "metabolic health". Boolean operators (AND, OR) were used. No restrictions were placed on language or publication date. Duplicate entries were removed using EndNote X20 software.

Study Selection

Two independent reviewers screened titles and abstracts based on eligibility criteria. Full texts were reviewed when necessary. Any disagreements were resolved through discussion and consensus. When consensus could not be reached, a third reviewer was consulted to make the final decision.

Data Extraction

Data extraction was performed using a standardized form developed for this review. Extracted information included:

Study authors and year

Population details (species, age, sex)

Exercise type, intensity, frequency, and duration

FOXO protein type (e.g., FOXO1, FOXO3)

Key findings related to FOXO activity and aging outcomes

The form ensured consistency across studies and reduced the risk of data extraction bias.

Quality Assessment

The risk of bias in animal studies was assessed using the SYRCLE risk of bias tool (Table 3). Human studies were evaluated with the Cochrane Collaboration's Risk of Bias 2.0 tool (Table 4). Studies were categorized as low, moderate, or high quality. Those assessed as low quality were excluded from the final synthesis.

Study Design: Experimental, observational, and interventional studies.

Table 1: Detailed Inclusion and Exclusion Criteria

Inclusion Criteria	Exclusion Criteria
Studies involving FOXO protein activation	Studies not assessing FOXO proteins
Exercise as primary intervention	Studies without an exercise component
Effects on aging, longevity, or related diseases	Unrelated outcomes or topics
Published original research	Reviews, commentaries, or letters

Information Sources and Search Strategy

Literature searches were conducted across the following databases: PubMed/Medline, Scopus and Web of Science. The search terms used included combinations of the following: "FOXO proteins", "exercise", "aging", "longevity", "FOXO3", "oxidative stress", and "metabolic health", applying Boolean operators (AND, OR). There were no restrictions on language or publication date. The following terms were used to search for articles: (FOXO OR FOXO1 OR FOXO3 OR FOXO4 OR "Forkhead box O") AND (longevity OR "life span" OR lifespan OR "life expectancy") AND (exercise OR "physical activity" OR "resistance training" OR "endurance training") AND (aging OR ageing OR senescence) AND ("metabolic health" OR "insulin sensitivity" OR "glucose metabolism" OR metabolism) No limits were set on the publication date and language. Duplicate articles were removed with the help of ENDNOTE X20 software.

Selection of Studies

Two independent reviewers evaluated study titles and abstracts based on the predetermined eligibility criteria. Disagreements were resolved through consensus or by consultation with a third reviewer. Studies meeting inclusion criteria underwent full-text review.

Data Extraction and Collection (Table 2)

Data extraction was carried out systematically, capturing:

- Study authors and year of publication
- Population details (species, age and gender)
- Type, intensity, frequency, and duration of exercise
- FOXO protein studied (mainly FOXO3)

Table 2: summarizes the extracted data from the included articles.

Author, Year	Population	Exercise Type	Frequency/ Duration	FOXO Protein	Outcomes
Williamson et al - 2010	85-year Women & 24-year Women	Progressive resistance Training	12 weeks/ 3*10 reps/ 70% 1 rm/3d, wk	Akt–$FoxO_{3a}$	After RE, both age groups showed dephosphorylation of cytosolic $FoxO_{3a}$, but older women could not activate nuclear FOXO3A to the same extent as younger women.
Wen et al – 2023	Drosophila/w1118 Fly/ 2 days old	Climbing test until fatigue	5 weeks	FOXO/PGC 1α/ SDH and FOXO/SOD	Exercise countered the age-related decline in muscle FOXO expression and enhanced longevity, particularly in FoxO-OE flies.
Jin et al – 2023	Drosophila/w1118 Wild male 2 days old	Endurance exercise Climbing speed / climbing endurance	6 days	Sirt1/FOXO/SOD	The research confirmed that muscle FoxO-specific overexpression (MFSO) and endurance exercise (EE) protect skeletal muscle (SM) and heart muscle (HM) from defects caused by a high-fat diet (HFD) by enhancing FOXO-related antioxidant pathways and lipid catabolism.
Zeng et al – 2020	Wistar rat 6-month -old male	Treadmill running / Resistance training/ ladder climbing Voluntary wheel running Treadmill and resistance	3 days a week /12 week	Akt/FoxO Signaling pathway	Mechanistically, exercise modulated the Akt/mTOR and Akt/FoxO3a signaling pathways, alongside activating AMPK, contributing to the beneficial effects on sarcopenia.
Fan et al – 2017	Four-month-old male Sprague-Dawley (SD) rats	swimming	swimming training at the exercise intensity of 45 min/d after 6 h of drug administration during 6-week exercise training period with 5 days training in each week.	AMPK – $FoxO_{3a}$ Signaling pathway	The research findings indicate that spermidine combined with exercise significantly mitigates D-gal-induced skeletal muscle atrophy in aging rats by enhancing autophagy and reducing apoptosis through the AMPK-$FoxO_{3a}$ signaling pathway.
Bedada et al- 2021	42 Participant With mild cognitive impairment	Treadmill	3-month and 6-month Bruse protocol	$FoxO_1$ expression	The research found that exercise training increased the expression of $FBXO_{32}$ and $FOXO_1$ in a gender-dependent manner among African Americans with mild cognitive impairment (MCI).
Woo et al – 2020	40 male C57BL/6 mice 38 -week-old and 20 male 58-week-old	Treadmill	5 times weekly / 8 weeks	$FoxO_1$	The study found that regular exercise training significantly increased levels of AMPK, SIRT1, $FoxO_1$, PGC-1α, and NAD^+ in the gastrocnemius muscles of middle-aged and aged mice compared to their respective control groups.

Quality Assessment

Quality and risk of bias were evaluated using established tools such as SYRCLE's risk of bias tool for animal studies (Table 3) and Cochrane Collaboration's risk of bias tool for clinical studies (Table 4). Studies were rated as high, moderate, or low quality. Low-quality studies were excluded from the synthesis to ensure robust findings. The SRYCLE strategy has validity and reproducibility. SYRCLE includes ten questions to assess the performance, friction, and bias of scientific articles included in the studies; the answers were scored with "Yes", indicating a low risk of bias, or "No", indicating a high risk of bias. Articles were evaluated as low, acceptable, or high quality. Those assessed as of low quality were excluded from the review.

Table3: This table presents the methodological quality of the included studies, assessed Individually using the SYRCLE instrument (Williamson, Raue, Slivka, & Trappe, 2010)

Author,Year	Q1		Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Q10	Score
Wen et al.,2023	?		Y	?	?	N	?	N	Y	Y	Y	6.0
Jin et al., 2023	?		Y	?	?	N	?	N	Y	Y	Y	6.0
Zeng et al.,2020	?		Y	?	?	N	?	N	Y	Y	Y	6.0
Fan et al,.2017	?		Y	?	?	N	?	N	Y	Y	Y	6.0
Woo et al,.2020	?		Y	?	?	N	?	N	Y	Y	Y	6.0

The study by (Williamson et al., 2010) was assessed using the Cochrane RoB 2.0 tool (Table 4) . The overall risk of bias was rated as "some concerns" due to the non-randomized design. While outcome measurements and adherence to the intervention were strong, the lack of randomization and allocation concealment introduced potential selection bias. Additionally, no pre-registered analysis plan was

available, raising concerns about selective reporting. (Bedada et al., 2021).

(Bedada et al., 2021) conducted a single-blind, controlled exercise trial in cognitively impaired older adults. The risk of bias was also judged as "some concerns", primarily because of incomplete outcome data—only half of participants had full gene expression data. While outcome measurement was robust (qRT-PCR) and blinding of assessors was applied, the absence of a detailed randomization method and lack of protocol registration may have introduced reporting bias (Table 4).

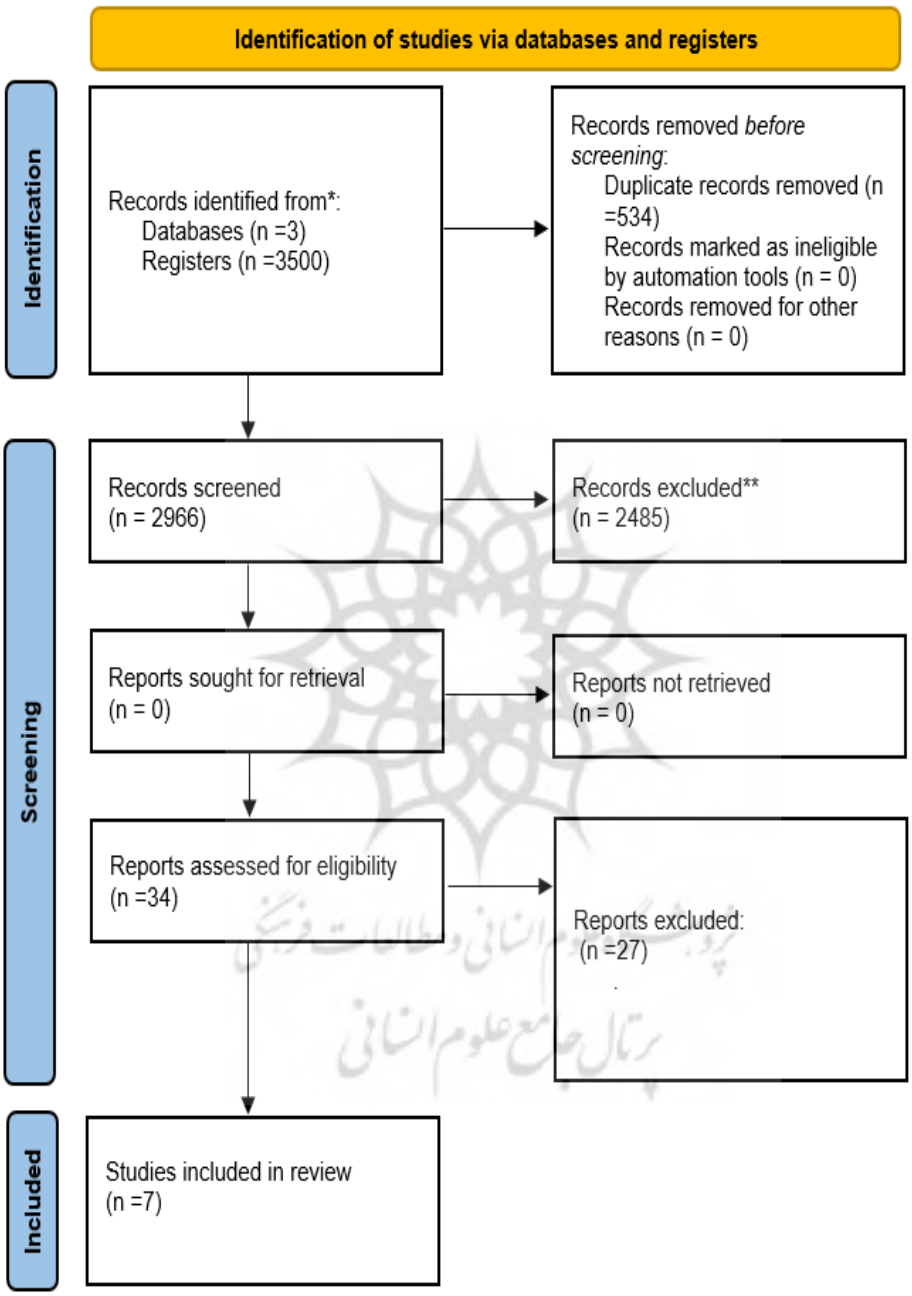
Table 4: Cochrane Collaboration's risk of bias

Domain	**Williamson et al. (2010)**	**Bedada et al. (2021)**
Randomization process	High risk	Some concerns
Deviations from intended interventions	Low risk	Low risk
Missing outcome data	Low risk	Some concerns
Measurement of the outcome	Low risk	Low risk
Selection of the reported result	Some concerns	Some concerns
Overall risk	Some concerns	Some cencerns

PRISMA Flowchart

The selection process of the articles included in this systematic review is detailed in the PRISMA flowchart (Figure 1).

Figure 1. PRISMA 2020 flow diagram illustrating the selection process of included studies.


Identification of studies via databases and registers
Identification
Records identified from*:
Databases (n =3)
Registers (n =3500)
Records removed *before screening*:
Duplicate records removed (n =534)
Records marked as ineligible by automation tools (n = 0)
Records removed for other reasons (n = 0)
Screening
Records screened
(n = 2966)
Records excluded**
(n = 2485)
Reports sought for retrieval
(n = 0)
Reports not retrieved
(n = 0)
Reports assessed for eligibility
(n =34)
Reports excluded:
(n =27)
Included
Studies included in review
(n =7)

Animal Studies

(Wen et al., 2023) investigated Drosophila melanogaster subjected to a climbing test protocol, revealing that endurance exercise enhanced FOXO/PGC 1α signaling and improved mitochondrial function. FOXO-overexpressing flies demonstrated a marked increase in muscle longevity and oxidative stress resilience. These findings align with the premise that exercise maintains muscular integrity and systemic health via FOXO-mediated antioxidant defenses.

In a complementary Drosophila study, (Jin, Wen, Chen, & Hou, 2023) focused on muscle FOXO-specific overexpression (MFSO) and endurance exercise under high-fat dietary stress. Their results confirmed that FOXO activation preserves muscle and cardiac tissue integrity through enhanced autophagy and lipid metabolism. Notably, SIRT1-FOXO-SOD signaling emerged as a crucial mechanism in mitigating diet-induced damage, emphasizing FOXO's multifaceted role in metabolic adaptation.

(Zeng et al., 2020) extended this investigation to Wistar rats, employing treadmill and ladder resistance training to examine FOXO3a modulation. Their study highlighted exercise-induced activation of the Akt/mTOR and Akt/FOXO pathways, which synergized with AMPK to suppress sarcopenia-related degeneration. The research provides direct mechanistic evidence linking FOXO activation with muscle maintenance and protein turnover in aging mammalian models.

(Fan et al., 2017) introduced a pharmacological dimension, combining spermidine with moderate swimming in aging Sprague-Dawley rats. Their data showed significant reductions in skeletal muscle apoptosis and enhanced autophagy, mediated via AMPK-FOXO3a signaling. The study reinforces FOXO's role in autophagy as a defense against age-associated cellular degeneration, bridging dietary and exercise-induced benefits.

(Woo & 2020)explored long-term treadmill training in middle-aged and aged C57BL/6 mice. The intervention upregulated FOXO1, PGC-1α, SIRT1, and NAD^+ in gastrocnemius muscle, indicating a systemic

rejuvenation of metabolic regulators. Their work supports a model where FOXO-dependent transcription cascades facilitate mitochondrial rejuvenation and stress adaptation in aging tissues.

Human Studies

(Williamson et al., 2010) conducted a controlled trial comparing young (24 years) and elderly (85 years) women undergoing resistance training. While both cohorts exhibited dephosphorylation of cytosolic FOXO3A, only the younger group achieved efficient nuclear translocation. This suggests that age-related impairments in FOXO signaling might dampen the full transcriptional benefits of exercise in older populations. (Bedada et al., 2021) examined African Americans with mild cognitive impairment and found that exercise intervention selectively upregulated FOXO1 and FBXO32, with gender-dependent effects. The use of qRT-PCR confirmed molecular fidelity, positioning FOXO1 as a neuroprotective agent responsive to behavioral interventions. This study provides human-based evidence linking FOXO expression with cognitive health and functional plasticity.

Collectively, these seven studies paint a compelling picture of FOXO transcription factors as central mediators of the physiological adaptations induced by physical exercise. Animal models consistently showed enhanced mitochondrial quality, autophagic flux, and oxidative stress resistance—primarily through FOXO3a-mediated signaling. The diversity of exercise modalities (treadmill, resistance, climbing and swimming) across species underscores FOXO's universal relevance in stress adaptation and energy homeostasis.

Human trials, though more limited in scope, underscore translational challenges. The diminished nuclear translocation in older women and the gender-specific expression in cognitively impaired individuals highlight the complexity of FOXO regulation in vivo. These findings suggest that age, sex, and baseline cognitive status may critically modulate FOXO responsiveness. Moreover, the discrepancy between cytosolic activity and nuclear efficacy calls for further investigation into FOXO's intracellular transport dynamics.

Taken together, these studies advocate for exercise as a powerful modulator of FOXO-dependent aging resistance. However, individualized exercise prescriptions may be required to optimize FOXO activation in humans, particularly in aged or neurologically compromised populations. Future research should prioritize longitudinal, mechanistic studies in humans to establish optimal intervention parameters and identify possible pharmacologic adjuncts to exercise-induced FOXO potentiation.

Exercise-stimulated FOXO3 activation

Physical exercise has long been known to have a positive effect on aging to success by overall stimulation of health and prevention of aging disease. Physical exercise has been shown to specifically activate FOXO3. Resistance training has also been found to induce FOXO3 in muscle cells and be correlated with enhanced metabolic activity and reduced oxidative stress (Williamson et al., 2010). FOXO3 caused by exercise is induced by exercise-induced induction of oxidative stress through several mechanisms involving one exercise-induced induction of the AMPK-SIRT1-FOXO pathway. Exercise-induced oxidative stress activates and enhances AMPK, which in turn activates SIRT1, exercise-induced deacetylase activation leading to the activation of FOXO3. It enhances mitochondrial function and thus age disease prevention and oxidative damage (Guan, Chen, & Dong, 2025; Jiang, Xu, Jiang, & Li, 2023)Exercise-induced exercise activation of FOXO3 is involved through several mechanisms.

In this regard, AMPK and SIRT1 pathways are activated since exercise-stimulated energy stress activates such pathways. AMPK phosphorylates SIRT1 in this process, which continues to carry out deacetylation and activation of FOXO3. The pathway leads to transcription of genes for genes involved in mitochondrial biogenesis, antioxidant defense, and metabolism regulation (Guan et al., 2025; Kim et al., 2021) Exercise is a potent intervention for healthy aging. Exercise leads to activation of FOXO proteins by diverse mechanisms:

1. Oxidative Stress and Antioxidant Pathways: Exercise is both a ROS generator and FOXO protein activator. FOXO proteins, in turn, enhance the antioxidant defense of the cell against oxidative stress by stimulating the activating antioxidant enzymes superoxide dismutase (SOD) and catalase (CAT) (Wen et al., 2023).
2. Energy sensing and Metabolism: Exercise controls energy-sensing pathways, i.e., AMPK and SIRT1, over FOXO proteins. Activation there of leads to FOXO-dependent gene expression towards mitochondrial biogenesis, fatty acid oxidation, and glucose metabolism (Woo & 2020; Zeng et al., 2020).
3. Autophagy and Mitochondrial Quality Control: Exercise enhances autophagy, a degradative cellular process for defective proteins and organelles. FoxO proteins are governed by modulating the transcription of primary autophagy genes such as Beclin1 and LC3. FoxO proteins regulate mitochondrial quality by controlling mitochondrial dynamics and biogenesis regulation (Fan et al., 2017; Zeng et al., 2020).

Exercise and FOXO in Specific Tissues

Exercise-specific function of exercise on FOXO proteins is most relevant to aging and longevity:
1. Skeletal Muscle: Exercise evokes activation of FOXO in skeletal muscle and therefore offers mitochondrial biogenesis, fatty acid oxidation, and autophagy. These activities are relevant in regulating the function and mass of the muscle during aging (Iijima, Ambrosio, & Matsui, 2025; Zeng et al., 2020)
2. Heart: Physical exercise will protect the heart from senescence-induced failure through activation of FOXO proteins that offer antioxidant protection and mitochondrial function enhancement. This reduces the incidence of cardiovascular senescence-related diseases (Wen et al., 2023)
3. Brain: Exercise increases FOXO activity in the brain and consequently causes neuroprotection against neurodegenerative disease and neuronal survival with DNA repair gene preservation, antioxidant defense, and apoptosis (Bedada et al., 2021; Torioge et al., 2022).

Exercise had protective functions against aging diseases and included cardiovascular disease, certain cancers, and neurodegenerative disease.

The Impact of FOXO3 Activation in Age-Related Disorders

In particular, exercise-induced improved insulin sensitivity and glucose metabolism has been linked with reduced risk for the onset of type II diabetes (Kim et al., 2021; Putra, Antarianto, & Hardiany, 2022) FOXO3 has been reported to suppress proliferation in cancer cells and trigger apoptosis. This would indicate that exercise-activated FOXO3 also has anti-cancer properties (Guan et al., 2025; Snarski, Ghimire, & Savkovic, 2024). In the appropriate setting, other research on neurodegenerative diseases like Alzheimer's has also indicated that activation of FOXO3 is followed by reduced beta-amyloid plaques causing disease and with greater neuron survival.

This would indicate that exercise, through FOXO3 activation, has some level of neuroprotection (Chang, He, & Xia, 2023; Guan et al., 2025) FOXO3 also plays a significant role in the regulation of many processes that require cell homeostasis and resistance to stress. FOXO3 has also been implicated in the prevention of age-related diseases including osteoporosis, cardiovascular disease, and neurodegenerative disease. For example, FOXO3 is employed in the regulation of bone-fat ratio in bone mesenchymal stem cells (BMSCs) and is protective against age-related bone loss. The relationship between FOXO3 and aging has been of particular concern to researchers, with several studies having made a prediction based on the function of FOXO3.

Genetic variation at the individual level has also shown some of the polymorphisms identified with longevity such as SNP FOXO3:rs2802292, which is associated with low risk for age-related disease (Klinpudtan et al., 2022; Silva-Sena et al., 2018; L. Sun et al., 2015) FOXO3 transcription factor holds the key to aging through its function in the regulation of many processes of stress resistance modulation, inflammation control, and activation of DNA repair mechanisms. FOXO3, for example, is associated with lower oxidative stress as well as amplified DNA repair mechanisms—two processes

pushed to an extreme degree in aging (Chang et al., 2023; X. Sun, Chen, & Wang, 2017). In addition, FOXO3 was revealed to inhibit pro-inflammatory pathways typical of aging through mechanisms of inhibiting inflammatory cytokines typical of aging (Torigoe et al., 2024; Torioge et al., 2022) Single nucleotide polymorphisms like the rs2802292 polymorphism of FOXO3 have been strongly correlated with human longevity. Longevity was associated with certain polymorphisms of FOXO3 in different populations, i.e., southern Chinese who were allele G carriers of single nucleotide polymorphism rs2802292.

Regulation of FOXO3 in human body

FOXO protein expression and function are controlled by genetic and epigenetic factors:

1. Genetic Variants: Certain genetic variants of FOXO proteins, i.e., FOXO3 longevity variant rs2802292, are responsible for human longevity. They enhance the activity of FOXO3, leading to better telomere maintenance and reduced inflammation (Torioge et al., 2022)
2. Epigenetic Modulations: Exercise induces epigenetic modulations such as histone acetylation and DNA methylation that result in the overexpression and activation of the FOXO protein. Modulation is responsible for the anti-aging effect of exercise (Lox, 2023; Sellami, Bragazzi, Prince, Denham, & Elrayess, 2021).

Human regulation of FOXO3 is complex with multiple pathways and post-translational modifications. One of the identified regulators of FOXO3 is the insulin/IGF-1 signaling pathway; under normal conditions when insulin/IGF-1 signaling takes place, FOXO3 gets phosphorylated and inhibited. Phosphorylation keeps FOXO3 retained in the cytoplasm and hence does not allow it to localize in the nucleus, where it would provoke the activation of target genes ((Kim et al., 2021).

In addition to phosphorylation, FOXO3 is also regulated by acetylation and ubiquitination. SIRT1, an NAD^+-dependent deacetylase, was found to catalyze deacetylation of FOXO3 and promote transcriptional

activation and nuclear translocation of FOXO3 (Guan et al., 2025; Kim et al., 2021)But ubiquitination leads to the degradation of FOXO3 and thereby suppresses its functional activity as well as controls cell growth and cell survival in the opposite direction (Snarski et al., 2024). FOXO3 is controlled by a wide variety of upstream signaling pathways with particular emphasis being placed on the insulin/IGF-1 pathway. FOXO3 phosphorylation induced by insulin/IGF-1 receptor activation inhibits its transcription activity by inhibiting nuclear entry (Kim et al., 2021) In contrast, exercise and caloric restriction therapy enhances the activation of FOXO3 by inhibiting insulin signaling to nuclear entry and transcription activity (Putra et al., 2022). Phosphorylation, acetylation, and ubiquitination are some of the shared post-translational modifications involved in the regulation of FOXO3 activity. Specifically, SIRT1 deacetylation is utilized to activate and translocate nuclei of FOXO3, while Akt phosphorylation is utilized to inhibit activity (Guan et al., 2025; Jiang et al., 2023). These PTMs are utilized to facilitate FOXO3 response to a large variety of cellular stressors and metabolic inputs. Interventions like caloric restriction and fasting activate FOXO3 by decreasing the circulating concentrations of circulating IGF-1 and activating the upstream kinases like AMPK (Putra et al., 2022; Williamson et al., 2010).Lifestyle interventions like exercise also include the majority of FOXO3 activation induction and thereby positioning its role in the context of health preservation in aging (Sheng, Lei, Yao, & Chao, 2023).

The Function of FOXO3 in Age-Related Diseases

Transcription factor FOXO3 has also been implicated in diseases of aging such as cardiovascular disease, cancer, and neurodegenerative disease. FOXO3's function in cardiovascular disease was also implicated to regulate genes needed for antioxidant and anti-inflammatory activity and therefore prevent atherosclerosis and cardiac failure (Chang et al., 2023; Kim et al., 2021). FOXO3 acts as a cancer tumor suppressor by inhibiting growth and triggering apoptosis but exhibits complex and context-dependent oncogenic activity (Snarski et

al., 2024). FOXO3 also functions to avoid beta-amyloid plaques deposition and neuron survival in neurodegenerative disorders like Alzheimer's disease. Thus, activation of FOXO3 has been proposed as a therapeutic application in the management of such a type of disease (Chang et al., 2023). FOXO3 in aging disease and longevity. FOXO3 in aging disease and longevity is established in the literature. Empirical research verifies that FOXO3 mediates aging through influencing some of the fundamental biological processes such as stress response, DNA repair mechanisms, and the regulation of inflammation, all of which form part of the aging process (Chang et al., 2023; Santos, Grenho, Martel, Ferreira, & Link, 2023; X. Sun et al., 2017).In addition, FOXO3 was discovered to repel age-related disease such as cardiovascular disease, cancer, and neurodegenerative disease by inducing antioxidant defense mechanisms and anti-inflammatory responses, anti-proliferation of cancer cells (Chang et al., 2023; Snarski et al., 2024).

Molecular Mechanisms Controlling Exercise and FOXO-Mediated Longevity

Exercise and FOXO-mediated longevity is controlled by many molecular mechanisms:

1.FOXO/PGC-1α/SDH Pathway: FOXO/PGC-1α/SDH pathway is a significant exercise-induced mitochondrial biogenesis and oxidative phosphorylation pathway. Exercise enhances mitochondrial function, mitigating oxidative damage and improving energy metabolism (Wen et al., 2023).

2.FOXO/SOD Pathway: FOXO/SOD pathway is an antioxidant defense pathway. The exercise-induced pathway enhances antioxidant enzyme expression and reduces oxidative damage (Wen et al., 2023).

3.AMPK-FOXO3a Pathway: Exercise activates the AMPK-FOXO3a pathway, and the pathway also plays a role in activating autophagy and inhibiting apoptosis. The pathway is most essential in skeletal muscle atrophy and aging (Fan et al., 2017; Zeng et al., 2020)

Exercise-stimulated FOXO protein activation is of the utmost significance in healthy aging and longevity:

1.Developmental Delaying Aging Disease: Exercise-induced FOXO activation postpones aging disease, such as sarcopenia, cardiovascular disease, and neurodegenerative disease, by enhancing cellular stress resistance and metabolic well-being (Izquierdo et al., 2025; Zeng et al., 2020).
2.Lengthening Extending Health span: Anti-aging function, through the FOXO protein, enhances extending health span, years lived healthfully and functional capacity (Izquierdo et al., 2025; Sellami et al., 2021).
3.Therapeutic Interventions: From molecular mechanisms to open the gate for therapeutic interventions towards healthy aging and prevention of age-related diseases by pairing exercise and FOXO activation (Lox, 2023; Putra et al., 2022).

Results

This review included seven studies—five in animal models and two in human populations—that examined the relationship between exercise and FOXO protein activation (Table 2) . Overall, consistent patterns emerged across species indicating that physical activity positively modulates FOXO signaling, particularly FOXO3, which is associated with enhanced mitochondrial function, antioxidant defense, and reduced markers of cellular aging.

Animal Studies

In animal models, a clear trend was observed: endurance and resistance exercises stimulated FOXO-related pathways. For instance, Drosophila studies by (Wen et al., 2023) and (Jin et al., 2023) demonstrated that muscle-specific FOXO overexpression, combined with exercise, enhanced mitochondrial health and prolonged lifespan, especially under metabolic stress. Similarly, (Zeng et al., 2020) and (Fan et al., 2017) found that treadmill running and swimming, respectively, activated AMPK-FOXO3a signaling in rats, contributing to muscle preservation and improved autophagy.
However, these studies varied in protocol intensity, duration, and biological models. For example, the effects observed in Drosophila may not fully translate to mammalian systems due to significant differences in physiology and gene expression regulation. Furthermore, many

animal studies had short intervention periods (typically under 12 weeks). limiting the ability to assess long-term effects.

Human Studies

In humans, the findings were more nuanced (Williamson et al., 2010) showed that while resistance exercise triggered FOXO3a dephosphorylation in both young and elderly women, only the younger group exhibited successful nuclear translocation of FOXO3a—a key step for gene activation. This suggests age-related impairments in FOXO signaling that may reduce exercise efficacy in older adults. (Bedada et al., 2021) investigated FOXO1 expression in African Americans with mild cognitive impairment. Their results showed exercise-induced FOXO1 activation, but with gender-dependent variability. While promising, the study was limited by its small sample size and incomplete gene expression data in nearly half the participants, affecting the reliability of its conclusions.

Critical Appraisal and Inconsistencies

Across studies, several limitations became evident. Sample sizes were often small, particularly in human trials, and intervention durations were relatively short. Methodological differences, such as varied exercise modalities and measurement techniques, introduced inconsistencies. Additionally, species-specific responses complicate the generalizability of animal data to human aging. While FOXO activation appears to be a common mechanism, the degree and nature of the response differ depending on age, sex, and baseline health status.

Discussion

Aging is driven by complex and interconnected biological pathways. Among them, the FOXO family of transcription factors—particularly FOXO3—has received increasing attention for its central role in promoting cellular resilience, regulating metabolism, and supporting longevity (Santos et al., 2023). This review highlights how physical exercise activates FOXO pathways, which in turn strengthen

antioxidant defenses, support mitochondrial function, and contribute to stress adaptation (Guan et al., 2025; Williamson et al., 2010).

Findings from animal models consistently show that both aerobic and resistance exercise can stimulate FOXO3 activity, reduce oxidative stress, and delay age-associated decline (Fan et al., 2017; Zeng et al., 2020). These studies emphasize FOXO proteins' role in mediating autophagy, mitochondrial biogenesis, and metabolic regulation. However, translating these mechanisms into human outcomes is complex.

In human studies, FOXO activation is also evident but appears to be influenced by factors such as age and sex. For example, older adults may show reduced nuclear translocation of FOXO proteins after exercise, limiting transcriptional activity (Williamson et al., 2010). Moreover, gender differences in FOXO1 activation (Bedada et al., 2021) suggest that personalized exercise prescriptions may be needed.

From a practical standpoint, these findings imply that moderate-intensity aerobic and resistance exercises may offer targeted anti-aging benefits—especially if designed to stimulate oxidative and metabolic pathways. For older adults and patients with chronic diseases, personalized programs based on genetic background, sex, and baseline metabolic health may optimize FOXO activation. Clinicians could integrate these findings into preventive health strategies aimed at maintaining physical function and delaying the onset of age-related diseases.

That said, the review also reveals several limitations in the available research. Many of the included studies—especially in human populations—suffer from small sample sizes, short intervention durations, and lack of long-term follow-up. In animal studies, results are often based on controlled environments and genetically homogeneous samples, which differ significantly from human physiology. Furthermore, methodological inconsistencies, such as variation in exercise type, intensity, and outcome measures, reduce comparability between studies.

Future research should prioritize longitudinal human trials that directly assess FOXO activation over time. Understanding how specific exercise parameters (e.g., frequency, intensity, duration) influence FOXO responses in diverse populations will be key to translating molecular findings into clinical applications.

Conclusion

Exercise is a powerful and accessible intervention for promoting healthy aging, in part due to its ability to activate FOXO transcription factors—particularly FOXO3. These proteins are central regulators of stress resistance, autophagy, mitochondrial function, and inflammation control, all of which contribute to cellular health and longevity. The evidence presented in this review supports the role of physical activity in enhancing FOXO-related pathways, with potential protective effects against age-related diseases such as cardiovascular disorders, cancer, and neurodegenerative conditions.

However, this review is not without limitations. Many of the included studies are preclinical and rely on animal models, which may not fully reflect human physiology. Human studies remain limited in sample size, duration, and diversity, and the exercise interventions reviewed varied widely in type and intensity, making direct comparisons challenging. These limitations restrict the generalizability of current findings and highlight the need for more standardized, long-term human research.

Despite these challenges, the findings offer meaningful implications for clinical practice and future research. Clinicians may consider incorporating moderate-intensity aerobic and resistance training as part of individualized care plans for older adults or patients at risk of age-related conditions. Researchers are encouraged to explore the dose-response relationship between exercise and FOXO activation in diverse human populations, and to identify biomarkers that can predict responsiveness to exercise interventions.

By deepening our understanding of the molecular mechanisms linking exercise to longevity, we move closer to designing targeted, evidence-

based strategies to preserve health, extend functional years, and improve quality of life during aging.

Funding:
This research received no external funding.

Institutional Review Board Statement:
Not applicable.

Informed Consent Statement:
Not applicable.

Acknowledgments:
The author wishes to thank Allameh Tabataba'i University for academic support and access to research databases.

Conflicts of Interest:
The author declares no conflict of interest.

ORCID

Ashrafsadat Anvarimanshadi	**https://orcid.org/**
Behzad Hajizadeh Maleki	**https://orcid.org/**
Nasrin Rafizadeh	**https://orcid.org/**
Davood Sabour	**https://orcid.org/**
Elnaz Safikhani	**https://orcid.org/**
Amirhossein Jafari	**https://orcid.org/**

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Corresponding Author: asanvary@gmail.com
How to Cite: Anvarimanshadi, A., Hajizadeh Maleki, B., Rafizadeh, N., Sabour, D., Safikhani, E., & Jafari, A. (2024). Exercise Reversing Aging Process and Inducing Longevity by FOXO Proteins Family, Journal of New Approaches in Exercise Physiology, 6(11), 151-180.
DOI: 10.22054/nass.2025.85287.1174.