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Reversal of age-associated frailty by controlled physical exercise: The pre-clinical and clinical evidences

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ABSTRACT

Demographic aging is one of the most serious challenges facing our society. Although we live longer, we do not live better because it is considered that approximately 16–20% of our life is spent in late-life morbidity. Older people have the greatest risk of developing frailty increasing the risk of presenting various adverse health events such as low quality of life, disability, hospitalization and even death. Frail men and women over 65 years old have lower muscle quality and muscle mass and higher percentage of body fat than non-frail people of the same age. In this review we will address the main physiological changes in the muscular and nervous system associated to aging. More specifically we will review the changes in muscle mass, quality, and strength relating them with the decrease in capillarization and muscular oxidative capacity as well as with the alterations in protein synthesis in the muscle with aging. The last section of the manuscript will be devoted to the animal models of frailty and the indexes developed to measure frailty in these models. We will finally address the importance of exercise training as an intervention to delay or even reverse frailty.

Introduction

The percentage of citizens over 65 years of age, in the world, is expected to increase from 18% to 28% by 2060; the percentage over 80 years will increase from 5% to 12% during the same period of time. This can be explained by advances in medicine and public health, changes in lifestyles, and improved education. The aging of the different physiological systems is not homogeneous. The skeletal muscle is one of those tissues that experience more dramatic and potentially more negative deterioration as we get older. For this reason, older people have the greatest risk of developing disabilities.

In 2019 sarcopenia has been defined as a muscular disease (muscle failure) caused by adverse muscle changes that accumulate throughout life.[1] Sarcopenia has its own diagnostic code: ICD-10-MC. The main determinant factor of sarcopenia is the decrease in muscle strength that underlies a low amount or muscle quality.[1] Therefore, the determination of muscle strength is considered the main factor to be measured to diagnose sarcopenia and to predict its adverse effects. The determination of muscle strength should be accompanied with the detection of low muscle quantity or quality for the confirmation of the diagnosis of sarcopenia. Finally, the severity of the disease is identified on the basis of physical performance tests such as the Short Physical Performance Battery (SPPB), Timed-up-and-go test (TUG) and/or 400 m walk. Although sarcopenia contributes to the development of the geriatric syndrome of frailty, both must be considered different entities. Sarcopenia is a disease, while frailty is a broader, complex, and multidimensional geriatric syndrome that encompasses not only the physical but also the cognitive and social dimensions.[2] Despite the ongoing controversy over an agreed definition of frailty, recent publications consider it a state characterized by a progressive decline of physiological systems related to aging, which results in a reduction of intrinsic capacity and confers extreme vulnerability to stressors, increasing the risk of presenting various adverse health events such as low quality of life, disability, hospitalization, and even death (Joint Action '724099/ADVANTAGE European Union's Health Program). The intrinsic capacity is considered the combination of all the physical and mental capacities that a person has and it includes psychological,

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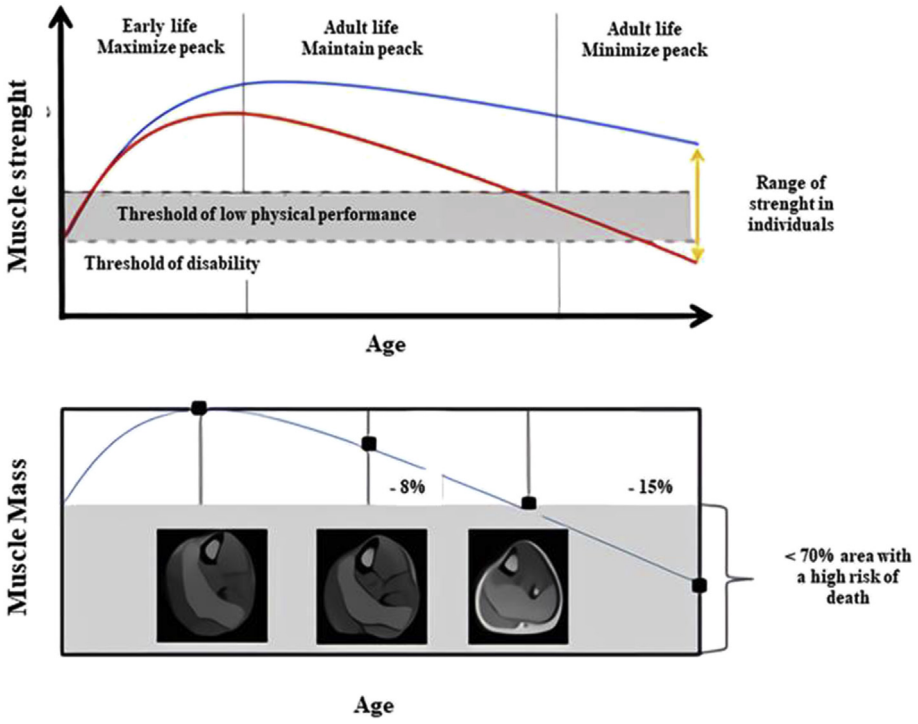

Fig. 1. Changes in muscle strength and muscle mass along lifespan.

cognitive, and functional aspects.[3]

The physical phenotype of frailty, described by Fried and colleagues[4], includes the following characteristics: Low gait speed (assessed by means of walking speed using sex and height corrected cut-off scores); weakness (defined as low grip strength corresponding to gender and body mass composition); poor endurance and energy, indicated by self-report of exhaustion; low physical activity (measured in kilocalories expended per week based on each participant report), and unintentional weight loss in prior year of the measurement.

It is considered as a frail person someone who scores 3 or more of these indicators. Pre-frailty is reached when a patient scores at least 2 of them and robustness when the score indicates 1 or none criteria. Even though Fried's criteria are the most common to evaluate frailty, there are other ways to achieve this goal.[5]

Frailty is very common with a global weighted prevalence of approximately 11% in people over 65 who live in the community. It has a clear impact on the costs of health services. Recent studies carried out in Germany, France, and Spain have determined their costs in the elderly both in the community and in hospitals. The incremental annual costs vary from 1,500 to 5,000 €/person depending on the state of frailty (pre-frail or frail) and the care environment (community or hospital).

Frail men and women over 65 years old have lower muscle mass and quality and higher percentage of body fat than non-frail people of the same age.[6,7] This is why we have included a section on the age-associated musculoskeletal changes with aging in our review.

The novelty of our review is that we bridge the gap between pre-clinical and clinical research by including a section on animal models of frailty. The biology of frailty is not well understood. This matches with the emergence of the development of experimental animal models for this geriatric syndrome. The pre-clinical models are based on "reverse-translation". Investigators have adapted the frailty scores originally developed in humans for its use in animal models. These models allow us the detection of the underlying mechanisms of frailty and are essential to explore intervention strategies in order to delay its onset or even to treat it. The translational objective of our review makes a difference with respect to the existing literature on this subject because we not only review the main animal models of frailty available for research but also the effects of exercise intervention on these models.

Searches were conducted in PubMed. Two independent search strategies were performed. One included the following terms: frailty, older people, and multiple expressions of exercise. In addition, reference lists from previous systematic reviews on exercise for the elderly were hand searched to identify trials on frail individuals. The other search strategy included term for frailty, rodent, mouse, rat, animal model, exercise, and pre-clinical research. Two review authors (C A-C and FM) independently screened the search results and performed data extraction.

Physiological changes in the muscular system associated to aging

Changes in muscle mass and strength/quality during aging

Muscle mass and strength vary throughout life, generally increasing with growth in youth, staying in middle-aged people, and decreasing with aging. In adulthood (up to ~ 40 years of age) it is considered that the highest levels are reached. After 50 years, there is a loss of mass (1–2% per year) and muscle strength (1.5–5% per year) of the legs[8] (See Fig. 1). As a result, many elderly people experience difficulties in carrying out activities of daily living and a significant increase in the risk of falls and fractures.[9] Falls are not only associated with morbidity and mortality in the elderly population, but are also linked to poorer general functioning and early admission to long-term care facilities.[10]

The loss of strength observed in the elderly is the result of muscle atrophy and alterations in the percentage of contractile tissue in the muscle cell.[8] The cross-sectional area of the skeletal muscle (CSA) decreases with age due to a reduction in the size of the muscle fibers, in the number, or in a combination of both.[11] The decrease in CSA is accompanied by structural changes in the muscles of the elderly (65–83 years of age). These contain less contractile tissue and more non-contractile tissue (fat and connective) compared to the skeletal muscle of younger people (26–44 years), which contributes to the reduction in the capacity of force production.[12] Muscular quality, defined as the force generated by each volumetric unit of muscle tissue, also decreases with age.[11] This measure

establishes the relationship between the strength of a certain muscle (dynamometry) and the area of the cross section of it. Measures of muscle quality also include the determination of fat infiltration or muscle attenuation.[12]

Low performance of the lower extremities is an important prognostic factor and predicts adverse health events in the elderly.[13] The change in the composition of muscle tissue in older people explains the reason why their muscle mass can be reduced to a greater extent than quantifiable through the measurement of the CSA. It also explains the reason why muscle strength is often reduced to a greater extent than muscle mass in the elderly.

At the cellular level, researchers have pointed to alterations in the sarcoplasmic reticulum as responsible for the reduction of muscle strength with age. The amount of Ca^{2+} released by the sarcoplasmic reticulum in response to depolarization is reduced in aged skeletal muscle due to a decoupling of the excitation-contraction process.[8] These data reinforce the importance of the measurement of muscle strength production to determine muscle aging. Another important aspect to consider in this context is the selective aging of certain types of muscle fibers and their relationship with atrophy and muscle dysfunction during aging. Most researchers agree that the size of type I fibers (slow-twitch) do not change substantially with age, while those of fast-twitch type II (IIa and IIx/d) do atrophy selectively.

Decreases in muscle capillarization and oxidative capacity during aging

In the skeletal muscle of older people, both the proportion of muscle fiber per capillary and its oxidative capacity are often reduced in comparison with the skeletal muscle of younger people.[14] Not only the number of capillaries and the area of the fiber are important for the oxygenation of the tissue, but also the way in which these capillaries are distributed. A heterogeneous distribution of capillaries has a negative impact on tissue oxygenation. The reduction in aerobic capacity during aging is related to the decrease in capillarity and also in muscle mitochondria. Our research group showed that one of the signaling pathways that are altered in skeletal muscle during aging is the p38 pathway, a mitogen-activated protein kinase, and the transcriptional coactivator PGC-1α, which promotes mitochondrial biogenesis.[15,16] In the young muscles we observed that, under different physiological stimuli, among which physical exercise was included, p38 modulated the mitochondrial biogenesis through the activation of PGC-1α, however this signaling pathway was altered in the skeletal muscle of old animals.[16] Reduced oxidative capacity due to mitochondrial dysfunction in skeletal muscle has been associated with the pathogenesis of sarcopenia and, finally, with the development of frailty.[17] An aged muscle will show damaged mitochondria and, on the contrary, young muscles will have healthy mitochondria that will be more resistant to deterioration and prevent the onset of frailty.

Aging of the nervous system and muscular atrophy

Another important aspect that is debated in the scientific literature is the relationship between the aging of the nervous system and that of skeletal muscle. The basal ganglia, involved in motor planning, suffer significant degenerative loss compared to other areas of the brain during aging, which compromises motor control.[12] This results in a reduction in physical activity and can create a cycle of inactivity leading to a loss of lean mass.[18] On the other hand, the size, total quantity and discharge rates of the motor units change with aging.[19] It is unknown whether these changes are the cause or the consequence of age-related reductions in the generation of strength and changes in muscle structure.[20] Studies that have investigated the number of motor neurons in the spinal cord and the number and size of motor axons in the anterior roots have shown that there is a loss of alpha motor neurons in the spinal cord, with the consequent degeneration of their axons while we age.[21]

On the other hand, it has been described that "orphan" muscle fibers are often reinnervated by some existing motor neuron through collateral innervation. Therefore, although there is a reduction in the number of motor units, some of them may increase in size.[8]

In addition to these changes in the morphology of the motor units, some researchers have shown that their activation rates decrease or gain variability with aging. The increase in variability has been attributed to a preferential denervation of type II fibers and their subsequent reinnervation with motoneurons associated with type I fibers. It can explain the deficiencies in motor control and in the production of strength in the elderly. The reinnervation of type II fibers with motor neurons of the adjacent type I fibers explains the increase in the co-expression of isoforms I and II of the myosin heavy chain in the muscle of older adults.[8] In addition to this, reductions in the myelination of the larger axonal fibers have been described with aging, which affects the conduction of the peripheral nerves and explains the characteristic slowness of the older adult.[21] All these modifications cause that in voluntary maximal muscle contractions there are significantly lower values of force than those that are achieved when the skeletal muscle is electrically stimulated during aging.[22]

Alterations in protein synthesis in muscle with aging

The decrease in the rate of protein synthesis is related to the decrease in muscle mass associated with aging. During aging, there is a deterioration in the anabolic response even with stimuli such as high protein diets or physical exercise.[23] Not all muscle proteins show altered synthesis rates. However, a reduction in the myosin heavy chains has been described with age and a correlation of this reduction with decrements in muscle mass and in the production of strength.[8] In fact, some authors have attributed the increase in the co-expression of the isoforms of the myosin heavy chain in the muscles of elderly people to an alteration in the protein synthesis. The mTORC1 complex plays a central role in the synthesis of muscle proteins through the activation of the S6K1 and 4E-BP1 proteins. This pathway is activated by a contractile stimulus and involves hormonal signaling.[24] In this cascade, the availability of amino acids is crucial and of the three branched chain amino acids (BCAAs), leucine plays a major role in the activation of the mTORC1 pathway.[25] In this sense an insufficient intake of proteins in the diet could contribute to the loss of muscle mass with age. However, defects in the S6K1 protein, lack of activation (phosphorylation), have also been described in the muscles of older adults in response to treatments with insulin or amino acids.[26] We enclose a summary of the main changes that occur in muscle tissue with aging (Fig. 2).

Animal models of frailty

The growing focus on frailty matches with the emergence of some experimental animal models, not only mice but also rats.[27,28] These models allow us the detection of potential biomarkers as well as the underlying mechanisms of frailty. Moreover, they are essential to explore intervention strategies in order to delay its onset or even to treat this geriatric syndrome.

The first experimental model of frailty was proposed in 2008[29] when a genetically altered mouse that did not express the anti-inflammatory cytokine Interleukin 10 was generated. This mouse model exhibits increased inflammation and strength decline consistent with human frailty. Another recent animal model for frailty is related to oxidative stress. The Sod1 deficient mouse exhibits changes in four of the five characteristics of human frailty proposed by Linda Fried.[4] Mice deficient in the NF-κB 1 subunit of the transcription factor NF-κB, show premature aging through increased inflammation and ROS mediated exacerbation of telomere dysfunction and cell senescence.[30] Two other potential interesting mouse frailty models could be the senescence accelerated prone mouse (SAMP)[31,32] or the prematurely aging mouse (PAM)[33] (See Table 1).

As in the clinical practice there are two main tools developed to assess

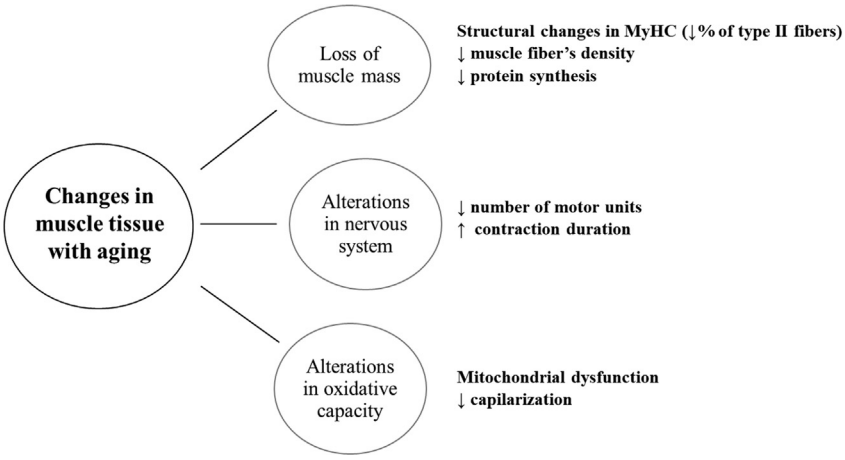

Fig. 2. Summary of the main changes in muscle tissue with aging.

frailty in mice[34]: the phenotype model[4] and the frailty index (FI).[5] The mouse FI assessment tool is based on the theory of the accumulation of health-related deficits through the quantification of functional in combination with biochemical measurements. Parks an co-workers[35] developed, for the first time, a mouse FI based on 31 health-related variables including activity levels, hemodynamic measurements, body composition, and metabolic status. The clinical basis for this model's development was the frailty index used in humans. Although it requires invasive procedures and specialized equipment, this evaluation cover a large range of physiological systems. This model was simplified, by the same research group, including only eight-item frailty parameters based on activity levels and weight changes.[36]

Thompson's research group created a clinically relevant frailty index for mice based on the clinical phenotype.[37] They proposed the identification of four frailty criteria (grip strength, walking speed, physical activity, and endurance) and provided cut-off points to assess frailty in the evaluated animals. They also published the mice neuromuscular health span scoring system, which included functional assessments as well as *in vitro* muscle contractility measurements.[38] Its invasiveness and time-consuming measurements are limitations of this model, but it has the advantage to show a reduced individual variability within groups.

More recently, our research group developed a new frailty score ("Valencia Score")[39] based on the human frailty phenotype.[4] In a longitudinal study, mice were evaluated from 17 to 28 months old and 5 criteria were assessed: unintentional weight loss; poor endurance; slowness; weakness and motor coordination. Using the "Valencia Score" we found that physical inactivity is a model of frailty in experimental animals because sedentary animals become frail as they age while lifelong spontaneous exercise significantly delays the onset of frailty. Moreover, our results show that by using the "Valencia Score" for frailty a prematurely aged phenotype can be identified even during the adulthood of animals. This opens up the possibility of carrying out preventive long-term interventions. Moreover, we have found that the individual frailty score of a given mouse at the late-adult, mature and old ages is shown to be a relevant predictor of its lifespan.[33]

Physical exercise as an intervention in pre-clinical and clinical models of frailty

The intervention strategies to avoid the loss of mass and/or muscle strength with age are three: nutritional, pharmacological, and physiological (training). It is important to note that these three strategies are related to each other, in fact, physical inactivity or a sedentary lifestyle lead to a rapid onset of anabolic resistance or a low protein synthesis

Table 1
Pre-clinical models of frailty.

Reference	Animal model	Assessments	Strengths	Limitations
[29]	IL-10 KO mice (C57BL/6 background)	Strength, activity, serum metabolites	Exploration of biological mechanisms of frailty	Based on inflammation processes Not natural aging
[35]	Mouse Frailty Index Male and female C57BL/6 mice	31 health-related variables (body composition, activity levels, hemodynamic measurements, metabolic status, etc.)	Large variety of health parameters Based on the "Frailty index" in humans	Invasive procedures and specialized equipment
[38]	NMHSS: neuromuscular mice healthspan scoring system Male C57BL/6 mice	Functional assessment (rotarod, grip strength) In vitro muscle contractility	Reduced individual variability within groups	Time-consuming and invasive procedures
[36]	Mouse clinical Frailty Index Male and female C57BL/6 mice	31 "clinical" items based on Parks et al., study	Non-invasive measures, fast and convenient	Excludes the cognitive aspect
[37]	Frailty phenotype index Male C57BL/6 mice	4 criteria: grip strength, rotarod, voluntary wheel running, endurance score	Non-invasive measures; Assessment of frailty with cut-off points	Time-consuming assessments
[39,33]	"Valencia score" for frailty Male C57BL/6 mice	4 criteria: weight loss, endurance, slowness, weakness and motor coordination	Non-invasive measurements Based on the Fried's frailty phenotype in humans	Specialized equipment required
[28]	Phenotypic frailty index for rats Male Fischer 344 rats	Battery of behavioral tasks (strength, speed, physical activity, endurance) based on the study of Liu et al.,	Correlation between the degree of frailty with survival	Consistent with frailty indices for humans and mice
[27]	Rat clinical Frailty Index Male Fischer 344 rats	Frailty index based on deficit accumulation (27 healthy-related deficits)	Includes the evaluation of various physiological systems	Excludes cognitive aspects and some functional performance

Table 2
Resistance training programs resulting in improvements in different functional parameters in old individuals.

Reference	*n*	Men	Women	Inclusion criteria	Duration in weeks	Main improvements
	27	5	22	TUG test	10	Muscle strength
[54,55]	28		28	Institunalized	48	Muscle strength
[56]	23			Functional daily difficulties	12	Muscle strength
[57]	32		32	Fried's frailty criteria	10	Muscle strength, muscle power, gait speed and TUG test
[58]	24			Nonagenarian	12	
[59]	51	20	31	Fried's frailty criteria	12	Fear of falling and self-reported physical function
[47]	117	54	63	SPPB test	24	8-Foot up and go test and endurance
[50]	151			Fried's frailty criteria	24	Fried's frailty criteria, gait speed and physical activity
[52]	40			Nonagenarian	8	1RM leg press

Table 3
Multicomponent exercise interventions for the treatment and prevention of frailty.

Reference	*n*	Men	Women	Inclusion criteria	Duration in weeks	Main improvements
[58]	24	7	17	Fried's frailty criteria	12	Muscle strength, gait speed, muscle mass and falls
[60]	51	20	31	Functional daily difficulties	12	Muscle strength and gait speed
[47]	117	54	63	SPPB test	24	Muscle strength and SPPB test
[48]	62		62	3 falls in the last year	12	TUG, sit-to-stand test and falls
[49]	616	280	336	Recurrent falls	12	Muscle strength and TUG test
[50]	246	95	151	Fried's frailty criteria	24	Muscle strength and gait speed
[51]	191	31	68	Katz Index	12	Muscle strength and gait speed
[52]	40	8	32	Institucionalized	8	Muscle strength, gait speed, and TUG test
[53]	69			Fried's frailty criteria	12	Muscle mass, muscle power, sit-to-stand test and SPPB test
[9]	100	46	56	Fried's frailty criteria	24	Fried's frailty criteria, Barthel and Lawton index, Tinetti, PPT and SPPB test

rate.[40]

As mentioned in the previous section we have seen that exercise is one of the most effective interventions in pre-clinical models of frailty.[39,41] Our results have been confirmed by other laboratories.[38] Recently, the benefits of HIIT (High Intensity Interval Training) on physical performance and frailty have been reported.[42] This study was performed in a cohort of 24 months-old mice, with an exercised group performing 3 sessions of HIIT (10 min duration) per week for 16 weeks. Significant improvements in grip strength, endurance, and gait speed were found in HIIT-trained mice that were associated with an increase in the mitochondrial biomass and related to a reverse in frailty.[42]

In the clinical practice it has also been shown that frailty can not only be delayed but also reversed by exercise training.[9]

Physical training has traditionally been divided into two categories: aerobic endurance training and resistance training. Aerobic training refers to exercise aimed at improving oxygen consumption ($\dot{V}O_{2max}$) or the time a person can maintain a physical activity at a certain intensity. The training against resistance refers to the exercise aimed at improving the maximum capacity for the generation of muscular strength. The adaptations induced by skeletal muscle training depend on the intensity, frequency, duration and type of exercise. The use of an appropriate exercise can delay or even reverse the physiological changes related to age that occur at the musculoskeletal level.[13,43]

Table 2 summarizes the main human studies in which old individuals, both institutionalized or community dwelling subjects, have followed a strength training programme resulting in improvements in different functional parameters.

Multicomponent interventions have also proved beneficial to treat frailty. Multicomponent exercise is defined as a program of endurance, strength, coordination, balance, and flexibility exercises, that have the potential to impact a variety of functional performance measurements. This type of exercise is a recommended alternative to more traditional exercise regimens, particularly due to its potential to impact functional performance outcomes in older adults (See Table 3).

Conclusions

There is a robust evidence showing that exercise training is an effective intervention to improve muscle function in the elderly and for the treatment and prevention of frailty. A common mistake in training with older adults is to consider that older people need to exercise moderately ("take it easy"). Although this may be true when starting an exercise program or in the presence of comorbidities (e.g., heart disease, diabetes, balance disorders), we now know that older people who are healthy respond to training in a manner similar to those younger. Two are the main challenges that we have to face in order to treat frailty with exercise. The first one is to consolidate these programs as a mandatory procedure in the clinical practice in frail and pre-frail individuals. The second challenge is to improve the exercise interventions. To issue a public recommendation of physical activity in the elderly is not an easy task because the changes with aging are not uniform and make the old population very heterogeneous. We should move towards the prescription of exercise as a precision medicine to look for an improvement on its effectiveness and in the adherence to the programs. We know that a multicomponent exercise intervention is the best choice available to improve frailty. However, tailored exercise interventions in terms of intensity and types of exercise, should be developed. Those interventions must be based not only in the functional status of the individual but should also take into account fall risk, co-morbidities, and nutritional aspects.

Conflict of interest

The authors declare no competing interests.

Submission statement

This manuscript has not been published and is not under consideration for publication elsewhere. If accepted, it will not be published elsewhere including electronically in the same form without the written consent of the copyright-holder.

Each authors' contributions

CA-C and FM performed the literature search and review and wrote the manuscript. AS-P, AGC, GO-G, ADlR, and AC analyzed and discussed the data and reviewed the manuscript. MCG-C and JV designed and supervised the review, secured funding, and wrote the manuscript. All

authors discussed the results, commented on and approved the last version of the manuscript.

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