

Effects of endurance, resistance and neuro-muscular electrical stimulation trainings to the anthropometric and functional mobility domains in elderly

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Background and aims. There's the need to increase physical activity engagement to promote healthy ageing. Different training protocols elicit different morpho-functional effects: the comprehension of the related assessment tests is a key to improve the specific proposals and to monitor adequately the adaptations. We aimed to identify the functional adaptation processes basing on different training protocols.

Methods. 40 healthy elderly (28 males and 12 females, 70.7 ± 4.39 y) were randomly divided into 4 groups: endurance, resistance, Neuro-Muscular Electrical Stimulation and control, trained for 12 weeks and ex-post evaluated on anthropometric and functional domains.

Results. We found: significant effect for gender, time and time \times protocols for Five Times Sit-to-Stand Test and Timed Up-and-Go test. Post-hoc analyses revealed effect for resistance and Neuro-Muscular Electrical Stimulation on Five Times Sit-to-Stand Test, and for endurance and Neuro-Muscular Electrical Stimulation on Timed Up-and-Go test. Correlations and factorial analysis linked Five Times Sit-to-Stand Test and Timed Up-and-Go test on the functional domain.

Conclusions. Medium-term physical interventions significantly modified functional characteristics of elderly. We found no ex-post effect on anthropometric parameters. The two functional tests are based on different underlying domains, our data therefore suggest to use both of them to specifically evaluate the training-induced functional adaptations in elderly. Our results promote the usefulness of evidence-based training.

Key words: Healthy ageing, Training, NMES, TUG, FTSST, Anthropometry

INTRODUCTION

Human ageing entails specific physiological modifications. Considering the current tendency towards an increase in the percentage of elderly population, the worthy focus is to understand deeply the course of healthy ageing: elderly need to avoid, inhibit or delay ageing-related chronic diseases, the loss of pro-activity and the loss of physical independence.

The engagement in physical activity is a cornerstone towards these aims, but the levels of participation are still low: one fourth of European population doesn't meet the minimum recommendations ¹, and the prevalence of physically inactive people drastically increases over 65 years-old age ².

The healthy effect of physical activity, facing with modern chronic disease is well known: the meta-analysis of Aune and colleagues shows that different subtypes of physical

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activity are strongly associated with a reduction in type 2 diabetes risk³. A recent work of Lachman and colleagues highlighted the protective effect of physical activity in older adults, even with low levels of engagement². In the healthy ageing paradigm, one of the key in later life is maintaining independence in the Activities of Daily Living (ADL), and different training protocols are effective in the attenuation of the disablement process⁴. Both endurance, resistance and Neuro-Muscular Electrical Stimulation (NMES) training protocols, if adequately proposed, showed their efficacy as Adapted Physical Activity (APA) for healthy elderly; however, different training protocols carry out the effects through specific physiological patterns, as demonstrated on neuro-motor level⁵, on circulating factors⁶, on transcriptional profile⁷, on regenerative system⁸.

Monitoring the effects of the proposed protocols is a need: the implementation of functional evaluation is the key to understand and direct specific training plans, both for research purpose and health promotion.

On this sight, Timed Up-and-Go test (TUG) and Five Times Sit-to-Stand test (FTSST, or 5STS, or FRSTST) have been widely using to evaluate functional mobility in older adults.

In particular, TUG test is strongly related to the mobility status of elderly⁹, and it is useful as a tool for prevention¹⁰. FTSST permits to identify poor exercise tolerance and impaired functional performances¹¹, and it is useful as an outcome measure in rehabilitation¹².

Chang and colleagues highlighted the usefulness of a physical performance battery for the early identification of frailty, among with the consideration of anthropometric parameters and taking into account the characteristics of the participants¹³. Lamb and Keene affirmed that the tests of physical capacity should be conducted under standardized environmental conditions, considering carefully the underlying construct and purposes¹⁴.

We aimed to understand the functional adaptations related to different training protocols, in order to define the specificity of functional parameters of motor behaviour in elderly, as measured by widely used TUG test and FTSST. We also aimed to understand how training-induced functional adaptations were related to anthropometric parameters. We finally investigated the effect on gender on these adaptations.

MATERIAL AND METHODS

PARTICIPANTS

Putting $\alpha = 0.05$ and $1-\beta = 0.8$, with an effect size of 0.3, the minimum number of participants was 36: we recruited 40 healthy subjects, in the age range of 62 to 82 years (Tab. I), reporting current physical inactivity during the initial interview. Exclusion criteria were: irregular ECG, osteo-articular pathologies, mild to medium cardio-circulatory pathologies, cancer, diabetes, non-controlled hypertension (diastolic pressure > 95 mmHg and/or systolic pressure > 150 mmHg), neurological or psychiatric disease, respiratory pathology, neuromuscular disease, genetic disease. All participants provided their written informed consent.

The study was conducted according to Helsinki declaration. The study was approved by the Ethics Committee of the University "G. d'Annunzio" of Chieti – Pescara, Italy (protocols no. 1634/07 COET and no. 1884/09 COET)

MEASURES

We measured the height and the body weight of the subjects, and calculated their Body Mass Index (BMI). We estimated the percentage of Fat Mass (%FM) using the plicometric method¹⁵.

Five Times Sit-to-Stand Test (FTSST) was applied according to a previous study¹⁶: at the signal of the evaluator, the subject stood up and sat down 5 times as quickly as possible, keeping the arms crossed on the chest. The evaluator provided preventively also the following instructions in order to standardize the execution: "Stand up fully and touch the chair in every repetition", "Do not touch your back to chair backrest". The subjects repeated the test three times and the best performance was considered.

Timed Up-and-Go (TUG) test was applied according to a previous study¹⁷: at the signal of the evaluator, the subject stood up from a chair, walked 3 m, turned beyond a ground line, walked back and sat down to the same chair. Two evaluators were positioned through the course to avoid any serious consequence in the case of a fall. The subjects repeated the test three times and the best performance was considered.

DESIGN AND PROCEDURES

We used an experimental longitudinal design to address the objectives. The subjects were divided randomly into

Table I. Participants. Descriptive characteristics of the recruited subjects; data are expressed as Mean \pm SD.

	Subjects	Age (years)	Weight (kg)	Height (m)
Overall	40	70.70 ± 4.39	72.10 ± 10.01	1.60 ± 0.08
Males	28	71.10 ± 5.05	75.90 ± 8.55	1.64 ± 0.07
Females	12	69.70 ± 2.02	69.70 ± 2.02	1.51 ± 0.03

four groups: endurance training ($n = 8$, 5 M and 3 F), resistance training ($n = 10$, 7 M and 3 F), NMES ($n = 10$, 7 M and 3 F) and control ($n = 12$, 9 M and 3 F). Initially, they were clustered in 4 groups with an equal number of participants, but due to logistical impairments 2 subjects were moved from endurance group to control. All the following tests were executed before (t0) and after (t1) the completion of three different training protocols, below detailed. For the control group we respected the same time of other groups, executing t1 analysis after 12 weeks from t0. Participants of the control group were required to report the current physical activity engagement at the end of the experimental time, in order to confirm the physical inactivity status, such as in t0; all of them confirmed the same status.

The training plans were set coherently with the general recommendations for the age: in particular, considering our group as older adults who were not active, we used a multiple-months progressive intervention, with a regular monitoring and a re-evaluation of the plans in accordance with functional changes¹⁸.

The endurance training consisted of three sessions per week, for a period of 12 week. The training plan was set in accordance with the minimum recommendation for vigorous-intensity aerobic activity in older adults of 20 min on three days each week¹⁸. The subjects were trained on a cycloergometer, pedalling at a constant intensity monitored through Heart Rate (HR) by a HR monitor (Polar Accurex Plus, Polar, Finland). HR training (HR_{tr}) was calculated according with Karvonen formula (19): $(HR_{max} - HR_{rest}) \times \text{Intensity} + HR_{rest}$. HR_{max} was derived by Tanaka formula²⁰: $208 - (0.7 \times \text{age-in-years})$. HR_{rest} was that one measured on ECG. Intensity was set at 0.6, 0.7 or 0.8 of HR_{tr} according to training plan. Intensity and duration were increased through the training period.

The resistance training consisted of three sessions per week, for a period of 12 week. The subjects were trained

on leg-press and leg-extension machines. Our plan met the minimum recommendation of two days each week for muscular strength in older adults, involving major muscle groups¹⁸. The subjects were trained with three series on leg-press and leg-extension machines. Intensity was based on the 1 Repetition Maximum (1RM) (Tab. II). The NMES protocol consisted of a training program that lasted 18 min, with 40 passive isometric bilateral contractions that were stimulated isometrically by a NMES device (Genesy 1200 Pro; Globus Srl, Codognè, Italy) for three sessions per week over a 12-week period. Pre-load and post-load phases were set identically to the other protocols. During the NMES session, the subjects were seating on a leg extension machine with the knee joint at 90°. Two active electrodes were positioned over the motor points of the quadriceps muscles, closely to the motor point of the Vastus lateralis and Vastus medialis muscles: the specific positions were identified starting from the reference tables of the products and searching for personalized locations. A dispersive electrode was placed 5 cm to 7 cm below the inguinal crease, to close the stimulation loop. The electrical currents followed these characteristics: rectangular-wave (75 Hz every 400 µs), rise time of 1.5 s, steady tetanic stimulation time of 4 s, fall time of 0.75 s, rest interval of 20 s between stimulations. The intensity was monitored throughout the time and it was gradually increased, keeping it close to the pain threshold of each subject, reaching at the end of the training period an intensity of 38 ± 9 mA.

STATISTICAL ANALYSIS

The calculations for statistical power were performed on G*Power software, version 3.1.9.3 (<http://www.gpower.hhu.de>). The statistical analyses were carried out using GraphPad Prism Software, version 7 (GraphPad Software, La Jolla, USA) and R-based open-source software Jamovi (<https://www.jamovi.com>).

Table II. Training protocols. Schedule and characteristics of the two training protocols; HRtr: Heart Rate training; 1RM: 1 Repetition Maximum; LP: Leg Press; LE: Leg Extension.

Protocol	Week	Intensity	Pre-load	Load	Post-load
Endurance	1-4	60-70% HR_{tr}	5' pedalling	30' pedalling	5' pedalling + 5' stretching
	5-8	60-70% HR_{tr}	5' pedalling	40' pedalling	5' pedalling + 5' stretching
	9-12	80% HR_{tr}	5' pedalling	40' pedalling	5' pedalling + 5' stretching
Resistance	1-4	60% 1RM	5' pedalling	3 series, 12 repetitions (both on LP and LE machine)	5' pedalling + 5' stretching
	5-8	70-75% 1RM	5' pedalling	3 series, 10 repetitions (both on LP and LE machine)	5' pedalling + 5' stretching
	9-12	80% 1RM	5' pedalling	3 series, 6-8 repetitions (both on LP and LE machine)	5' pedalling + 5' stretching

org). Identification of outliers was performed with ROUT method ($Q = 1\%$). Normality of the distributions was assessed with D'Agostino and Pearson omnibus test. Equality of variances was assessed with Levene's test. Data are reported as Mean (M) \pm Standard Deviation (SD). Three-way ANOVA for Repeated Measures was used to analyse differences between groups, gender and over the two time spots, in addition to Tukey (for time \times gender significance), and Sidak (for ex-post significance of each protocol) correction for multiple comparisons. We used correlation matrixes with Pearson method to test the correlations between parameters at t0 and t1. We ran Principal Component Analysis (PCA) analysis to evaluate the factorial relation between parameters (Oblimin rotation, number of components based on eigenvalues > 1 and scree plot). Significance was indicated as * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$. Effect size was reported with partial η^2 . Given α , effect size and sample size, we calculated power ($1-\beta$).

RESULTS

All the participants performed TUG test, both at t0 and t1, with a better time than the cut-off value for normal mobility proposed by Bischoff and colleagues⁹. There were no significant differences in BMI. There was a tendency for the interaction effect (time \times protocol, $p = 0.068$). Post-hoc analyses revealed no significant differences (Fig. 1).

There was a significant effect for gender in the %FM ($p < 0.001$, partial $\eta^2=0.724$, $1-\beta=0.769$), and a

tendency for time effect ($p = 0.08$). Post-hoc analyses revealed females to have greater values, both at t0 ($p < 0.001$) and t1 ($p < 0.001$) (Fig. 2).

We found a significant reduction from t0 to t1 in FTSST values, with the effects of time ($p < 0.001$, partial $\eta^2 = 0.505$, $1-\beta = 0.193$), time \times protocol ($p = 0.030$, partial $\eta^2 = 0.241$, $1-\beta = 0.537$) and a tendency for protocol ($p = 0.064$). There was a gender effect ($p = 0.009$, partial $\eta^2 = 0.196$, $1-\beta = 0.231$, females have greater values than males). Post-hoc analyses revealed independent negative effect for resistance ($p = 0.035$), NMES ($p < 0.001$), males ($p < 0.001$) and females ($p = 0.003$) (Fig. 3). It is worth noting that a negative effect on this test means a functional improvement. We found a significant reduction from t0 to t1 in TUG values, with the effects of time ($p < 0.001$, partial $\eta^2 = 0.373$, $1-\beta = 0.219$), time \times protocol ($p = 0.040$, partial $\eta^2 = 0.232$, $1-\beta = 0.563$). There was a gender effect ($p = 0.003$, partial $\eta^2 = 0.254$, $1-\beta = 0.191$, females have greater values than males). Post-hoc analyses revealed independent negative effect for endurance ($p = 0.037$), NMES ($p < 0.001$) and males ($p = 0.002$) (Fig. 4). It is worth noting that a negative effect on this test means a functional improvement.

Correlation matrixes (Tab. III) showed a good correlation between FTSST and TUG, and a light correlation between %FM and the two afore-mentioned tests. Factorial analysis (Tab. IV) demonstrated the association of TUG and FTSST into the same functional domain, and the anthropometric domain to be separated into two different components. Thus, we identified the following three domains: Functional, Anthropometric BMI,

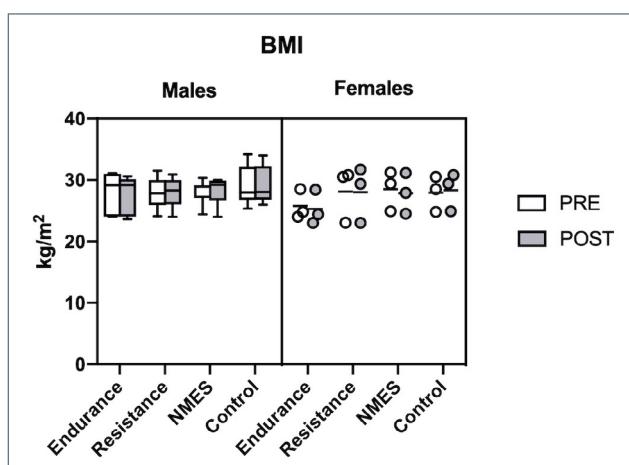


Figure 1. BMI. Results of the Body Mass Index calculations; white boxes and dots represent t0 values, grey boxes and dots represent t1 values; horizontal lines represent Mean; whiskers represent min to max; vertical line divides male and female data; NMES: Neuro-Muscular Electrical Stimulation; M: Males; F: Females.

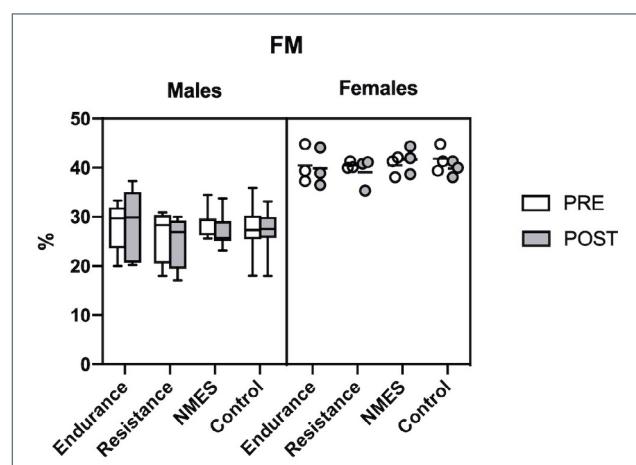


Figure 2. FM. Results of the percentage of Fat Mass estimation from plicometry; white boxes and dots represent t0 values, grey boxes and dots represent t1 values; horizontal lines represent Mean; whiskers represent min to max; vertical line divides male and female data; NMES: Neuro-Muscular Electrical Stimulation; M: Males; F: Females.

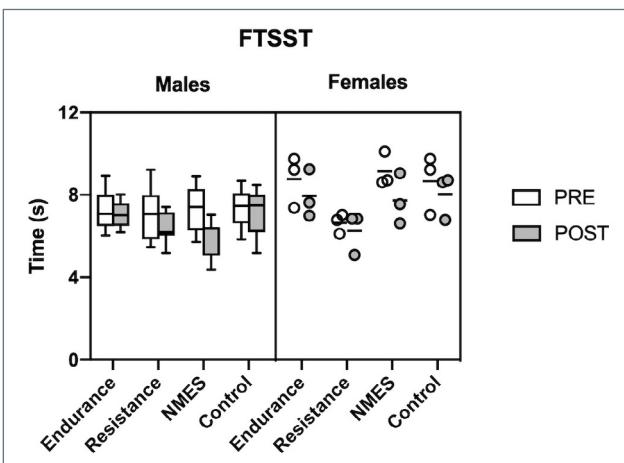


Figure 3. FTSST. Results of the Five Times Sit-to-Stand Test; white boxes and dots represent t0 values, grey boxes and dots represent t1 values; horizontal lines represent Mean; whiskers represent min to max; vertical line divides male and female data; NMES: Neuro-Muscular Electrical Stimulation; M: Males; F: Females.

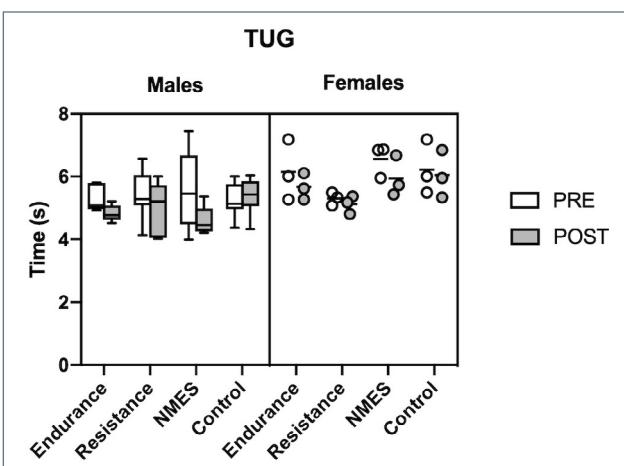


Figure 4. TUG. Results of the Timed Up-and-Go test; white boxes and dots represent t0 values, grey boxes and dots represent t1 values; horizontal lines represent Mean; whiskers represent min to max; vertical line divides male and female data; NMES: Neuro-Muscular Electrical Stimulation; M: Males; F: Females.

Anthropometric FM. Component correlations revealed a higher value in the comparison between Functional and Anthropometric FM, rather than between Functional and Anthropometric BMI.

DISCUSSION

Our recruited subjects demonstrated a good accomplishment and satisfaction on the course of study and on the perceived outcomes after training protocols. We did not find any significant difference in the

Table III. Correlations. Results of correlations (r values) test before (t0) and after (t1) the training period; BMI: Body Mass Index; %FM: percentage of Fat Mass; FTSST: Five Times Sit-to-Stand Test; TUG: Timed Up-and-Go test; * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

t0	BMI	%FM	FTSST
%FM	0.192		
FTSST	0.135	0.389*	
TUG	0.120	0.374*	0.667***
t1	BMI	%FM	FTSST
%FM	0.249		
FTSST	0.209	0.452**	
TUG	0.204	0.496**	0.509**

Table IV. PCA. Principal component Analysis (KMO test for sampling adequacy 0.710) with all the parameters; the first table shows the component loadings, the second table shows the component summary, the third table shows the component correlations.

	Component		
	1	2	3
BMI t0		0.991	
%FM t0			1.016
FTSST t0	0.888		
TUG t0	0.892		
BMI t1		0.999	
%FM t1			0.947
FTSST t1	0.843		
TUG t1	0.628		

Component	% of variance	Cumulative
1	34.3	34.3
2	25.4	59.7
3	25.1	84.7

	2	3
1	0.202	0.474
2		0.240

anthropometric characteristics from t0 to t1. Our results agree with that ones of Binns and colleagues, who reported no significant difference on Lean body Mass (LM) after 20 week (2 session/w) of High Velocity Resistance Training in healthy old subjects (77 ± 6.4 y), both in males and in females²¹. Thus, on healthy elderly, medium-term physical interventions are unable to significantly improve anthropometric characteristic. We should promote the long-term physical exercise engagement to see beneficial effects on anthropometric parameters of healthy older adults²².

Females have greater percentage of FM respect than males: other than the well-known hormonal traits, Tian and colleagues reported the same trend of age-related changes in body composition in a large cohort, with females having higher FM across the lifespan²³. The course of anthropometric adaptations improves the need to promote a long-term involvement in physical activity, beyond the duration of research protocol, in order to provide preventive morphological adaptations in healthy elderly. Surprisingly, factorial analyses revealed BMI values and FM values to be separated in the latent components. Furthermore, component correlations of factorial analysis and correlation matrixes revealed a higher correlation of functional domain with the domain of FM. Therefore, we should take into account which anthropometric domain has to be considered in the links to functional adaptations.

We found a significant interaction (time × protocol) effect on both FTSST and TUG: different training protocol entail different effects. We expected that elderly trained with NMES and RT were more responsive on functional tests, especially on FTSST: surprisingly, we found similar tendencies for the effects on each group. This result could be explained considering that we found TUG and FTSST to be correlated. The high correlation support the idea of a functional articulated domain. Goldberg and colleagues also reported a correlation between the two afore-mentioned tests, suggesting the usefulness of FTSST as a measure of functional mobility in older adults²⁴. Interestingly, we found NMES group to be significantly responsive to both TUG and FTSST.

However, the low achieved power, due to high Coefficient of Variations (CVs) and limited number of participants in sub-groups, suggest us to take cautiously into account these results.

In particular, considering the independent effect of resistance and NMES on FTSST values, we consider this test more related to the local strength respect than TUG. This interpretation is in agreement with that one by Bohannon and colleagues, who highlight the relation of FTSST with lower limb strength; moreover, these authors found no correlation between FTSST and anthropometric profile²⁵.

Supporting the difference in functional sub-domain measure, we found independent effect of endurance and NMES, but not resistance, on TUG values: as affirmed by Maden-Wilkinson and colleagues, the changes in muscular mass and strength only partially explain the impairment in functional performance²⁶.

Considering the local increment of maximal isometric strength, as reported in previous study with the same protocol⁸, we expected to find a significance for NMES only on FTSST. Surprisingly, we found NMES to be effective on both the functional tests. Although we did not attempt

to determine the mechanisms, we may suggest these results to be related to a maintenance in neuroplasticity²⁷. As expected, we found gender differences in functional test (males exerted better performances), but not with an interaction effect with time and protocol. Ibrahim and colleagues affirmed that functional pattern course through ageing may be similar across genders²⁸. In our data, the number of females did not permit us to achieve adequate power in statistical comparisons of the functional tests, thus this limit did not permit us to propose a clear framework of gender-related adaptations: other studies with a large number of participants are required to go further in this topic.

FTSST and TUG are widely used tests to assess functional mobility in elderly: in our study, PCA and correlation matrixes revealed that there's a common base: however, our results revealed FTSST to be more related by a vigour performance and TUG more related to a more articulated motor behavior, as highlighted by the post-hoc effect of endurance training. Thus, we suggest the use of both of them to better characterize the course of functional adaptations in elderly.

Concerning the timeline of the evaluation, starting from the present findings, we suggest a multi-spot comprehensive interpretation with a sequential evaluation protocol, such as that one proposed by our group for the analysis of Maximal Voluntary Contraction (MVC) at t0, t4, t8, t12 e t28 (after 16 weeks of de-conditioning)⁷. On the basis of recent results, demonstrating MVPA (Moderate-to-Vigorous Physical Activity) as a strong predictor for TUG and FTSST in healthy elderly²⁹, we should include the assessment of specific physical activity engagement in ex-post studies to improve the comprehension of multi-domain effect of specific exercise.

Considering our results, we suggest further studies with similar designs in order to go further in the identification of exercise-related adaptations, clustered by gender and age. Our results can be a base to suggest effective training protocols in healthy elderly.

In perspective, we could address other functional domain in elderly, such as balance³⁰, and the physiological adaptive main pathways implied in ageing³¹. We recently demonstrated a specific cross-talk between lower and upper limbs, the Bottom-Up Rise Strength Transfer (BURST), mainly linked to endurance training: we may therefore move forward in the comprehension of physiological mediators of training-related functional improvements³².

Considering the differences on the effect between training protocols and the evidence of gender differences, we should carry on towards an evidence-based physical activity training plans.

In conclusion, the results of the present study highlight and specify the effects of different trainings in elderly in

a functional mobility framework, brickling a piece in order to better promote adequate physical activity-based strategies towards an active and healthy ageing.

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CONFLICT OF INTEREST

The Authors declare to have no conflict of interest.

PUBLICATION STATEMENT

This manuscript has not been published elsewhere and it is not under consideration for publication elsewhere. Its publication is approved by all authors.

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