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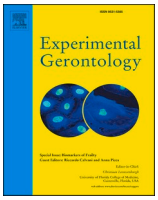

Are inflammatory markers associated with sarcopenia-related traits in older adults with sarcopenia? – A cross-sectional analysis of the ENHANce study

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ABSTRACT

Aims: To explore the relationship between inflammatory markers and sarcopenia-related traits in sarcopenic older adults.

Methods: Baseline data of the ongoing Exercise and Nutrition for Healthy AgeiNg (ENHANce) study were used for a secondary, exploratory, cross-sectional analysis. ENHANce is a 5-armed triple blinded randomized controlled trial, in older adults (>65y) with sarcopenia defined according to the revised criteria of the European Working Group of Sarcopenia in Older People (EWGSOP2) aiming to assess the effect of combined anabolic interventions (protein supplement, omega-3 supplement and physical exercise) on physical performance, compared to single/placebo interventions. Inflammatory markers C-reactive protein (hs-CRP), albumin, interleukin-1β (IL-1β), IL-6, IL-8, and tumour necrosis factor-α (TNF-α) were assessed at baseline. Spearman's rho (ρ) correlation coefficients were calculated to associate these inflammatory markers with baseline sarcopenia-defining parameters (handgrip strength, chair stand test, appendicular lean mass [aLM], gait speed, Short Physical Performance Battery), physical activity (step count) and quality of life (SF-36, SarQoL).

Results: We included 40 sarcopenic subjects (15 men/25 women, age 77.1 ± 6.8 years). Contrary to expectations, the pro-inflammatory IL-1β correlated positively with handgrip strength (ρ: 0.376; $p = 0.024$) and IL-6 with aLM (ρ: 0.334; $p = 0.0433$). IL-6 inversely correlated with step count (ρ:-0.358; $p = 0.048$). Subgroup analysis revealed important gender differences. IL-8 inversely correlated with handgrip strength in women (ρ: -0.425; $p = 0.034$) but not in men. In contrast, pro-inflammatory cytokines CRP (ρ: -0.615; $p = 0.019$), IL-6 (ρ: -0.604; $p = 0.029$) and TNF-α (ρ: -0.615; $p = 0.025$) inversely correlated with the SF-36 physical component score in men but not in women.

Conclusion: Although Inflammageing might play a role in sarcopenia-related traits, this exploratory study highlights an important role of gender. Future research should take this into account when elucidating the Inflammageing-sarcopenia interplay.

1. Introduction

Sarcopenia is a muscle disease, characterized by loss of muscle mass and function, leading to 'muscle failure' (Cruz-Jentoft et al., 2019). Primary sarcopenia is age-driven and is a growing concern in the ageing population. One of the major mechanisms behind the onset and progression of sarcopenia is the chronic low grade inflammatory state related with ageing, the so-called 'Inflammageing' (Dalle et al., 2017). This concept was originally introduced by Franceschi et al. in 2000 and is presumed to play a major role in several age-related diseases (e.g., dementia, cardiovascular disorders or osteoarthritis) (Franceschi et al., 2000). With ageing, macrophage activation increases, which contributes

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to a chronic subclinical inflammatory process in older adults, portrayed by increased pro-inflammatory cytokines and decreased anti-inflammatory cytokines (Franceschi et al., 2000).

Several cytokines are involved in inflammageing and the exact way in which they are entwined with sarcopenia urges further unravelling (Bano et al., 2017; Schaap et al., 2006; Schaap et al., 2009). In the context of sarcopenia, a lot of attention goes to interleukin-6 (IL-6) (Pan et al., 2021). This cytokine is also a myokine, thus both produced by the muscle as well as other cell types (Pedersen and Fischer, 2007). IL-6 can play a dual role depending on the levels and duration of exposure. On the one hand, low levels of IL-6 can stimulate satellite cell activation and regeneration of myotubes (therefore beneficial for the muscle), on the other hand chronically high increased levels of IL-6 promote skeletal muscle wasting through altering the metabolism of lipids and proteins, as well as impairing myogenic differentiation (Belizário et al., 2016). Accordingly, previous findings regarding IL-6 and sarcopenia are contradictory. A study of Schaap et al. demonstrated that high levels of IL-6 are predictive for muscle strength loss (Schaap et al., 2006), whereas a recent study of Liu et al. in $n = 77$ community-dwelling older adults could not find an association with sarcopenia, defined according to the Asian Working Group of Sarcopenia (AWGS) (Liu et al., 2021). Similarly, a systematic review found no associations between sarcopenia (defined according to various definitions) and IL-6 (Bano et al., 2017). However, gender differences may play a role in the Inflammageing-sarcopenia interplay as a recent meta-analysis suggested that IL-6 might have different cut-off levels in men than women to predict sarcopenia, since men with better muscle condition had higher levels of IL-6 in their plasma than women with worse muscle parameters (Mikó et al., 2018). Similarly, Marzetti et al. demonstrated that physical frailty and sarcopenia (PF&S, defined on a combined presence of physical frailty and low muscle mass) might be characterized by an inflammatory profile with increased levels of P-selectin, C-reactive protein (CRP) and interferon γ-induced protein 10, and gender-specific patterns of this inflammatory profile might be applicable in older adults with PF&S (Marzetti et al., 2019).

Besides IL-6, various other cytokines and inflammatory markers are suggested to be linked with sarcopenia. For example, CRP levels might be associated with the presence of sarcopenia (defined using various definitions) (Bano et al., 2017). Accordingly, a recent meta-analysis confirmed the association of increased CRP with low muscle strength (Shokri-Mashhadi et al., 2021). Furthermore, tumour necrosis factor α (TNF-α) is suggested to influence sarcopenia onset and progression (Wang et al., 2018). Regarding interleukin-8 (IL-8), data are conflicting. Whereas one study demonstrated that higher IL-8 levels were associated with lower appendicular lean mass (aLM) and an increased risk for sarcopenia in $n = 336$ community-dwelling older adults (Westbury et al., 2018), another found IL-8 to be lower older adults with PF&S (Marzetti et al., 2019).

As mentioned before, gender differences can influence the relationship between inflammatory markers and sarcopenia. However, contradictory findings might also be related to the differences in the definitions used to diagnose sarcopenia, as laboratory biomarkers can vary in persons with sarcopenia according to varying definitions (Lampignano et al., 2021). In 2019, the European Working Group on Sarcopenia in Older People revised their diagnostic criteria (EWGSOP2), placing muscle strength upfront as the primary determinant, instead of muscle mass (Cruz-Jentoft et al., 2019). Data on inflammatory markers in older adults with sarcopenia according to the EWGSOP2 are scarce (Liu et al., 2021) and lack an exploration of associations with sarcopenia-related traits, which might be useful to shed new light on this web of conflicting findings.

Therefore, the objective of present study was to explore the levels of inflammatory markers (CRP, albumin, IL-1β, IL-6, IL-8 and TNF-α) in older adults with sarcopenia according to the EWGSOP2 definition. Moreover, we aimed to explore associations of these inflammatory markers with various sarcopenia-related traits, like the individual sarcopenia-defining parameters (muscle strength, mass, and physical performance), as well as physical activity and quality of life in these older adults with sarcopenia. We hypothesized that pro-inflammatory markers are negatively associated with sarcopenia-related traits.

2. Methods

2.1. Subjects and study design

The present study was an exploratory, secondary, cross-sectional analysis of baseline data from the ongoing Exercise and Nutrition for Healthy AgeiNg study (ENHANce) (Dedeyne et al., 2020). Older adults (≥65y) with sarcopenia were recruited for ENHANce in the University Hospitals Leuven (UZ Leuven), Belgium. ENHANce aims to assess the effect of combined anabolic interventions (protein supplement, omega-3 supplement and physical exercise) on physical performance in older adults with sarcopenia, compared to single interventions or placebo in a 5-armed triple-blinded randomized controlled trial (RCT). Details of the methodology have been published before (Dedeyne et al., 2020). To summarize, community-dwelling or assisted living older adults (≥65y) were recruited to participate in a 12-week intervention, followed by 12-weeks of follow-up. In case one of the following was present, subjects were excluded: not able to communicate in Dutch, English or French; allergy to milk, soy, peanut or peanut oil; Mini-Mental State Examination (MMSE) score < 21 (Folstein et al., 1975); terminal illness with a prognosis <6 months; a protein intake higher than 1.5 g/kg body weight (BW)/day; participation in a training program more or equal than twice per week for the last 6 months; active uncontrolled disease, acute cardiovascular problems or negative advise of doctor to perform physical activities; 25-hydroxyvitamin D blood concentration <20 ng/L; glomerular filtration rate <30 mL/min/1.73 m^2; fasting glycaemia >126 mg/dL; usage of anti-diabetic medication; presence of impairments or diseases that impose problems to study participation. Muscle strength status was determined at screening visit, followed by a baseline visit with blood sampling and assessment of the various sarcopenia-related traits. Participants for the present study were included from February 2018 until December 2021. The study was registered at ClinicalTrials.gov (NCT03649698). Ethical approval was obtained by the UZ/KU Leuven Ethical committee (s60763). All subjects provided written informed consent. The study was reported according to the STROBE checklist for cross-sectional studies (von Elm et al., 2007), see Supplementary Table S1.

2.2. Sarcopenia-defining parameters

Muscle strength was measured through an evaluation of the grip strength with the Jamar 1 hand-held dynamometer (TEC Inc., Clifton, NJ, USA). Maximal grip strength was recorded as the highest of three measurements at both sides (Roberts et al., 2011). Lower extremity muscle strength was assessed through the chair stand test (Cruz-Jentoft et al., 2019), by timing the duration that a participant needed to stand up and sit again for five times in a row. To assess muscle mass, appendicular lean mass (aLM) was measured with a whole-body Dual-energy X-ray absorptiometry (DXA) scan on a QDR 4500A Discovery scanner (Hologic Inc., Bedford, MA, USA). Scans were analysed using Hologic APEX 4.0 software. To assess physical performance, gait speed (m/s) was used. The subject was instructed to walk six meters at usual pace. The time was measured over four meters, i.e. starting from the moment the participant's foot passed the mark of the one meter line until the foot passed the mark of the five meter line according to the British Columbia (BC) guidelines (BCGuidelines.ca, 2017). As an additional measure of physical performance, the Short Physical Performance Battery (SPPB) was used. SPPB is a multicomponent test combining a chair stand test, a gait speed test and a balance test. Each individual domain is scored from 0 to 4 and the total score ranges from 0 to 12 points, with a score of 12 indicating most optimal physical performance (Guralnik et al., 1994).

2.3. Sarcopenia definition

Sarcopenia was defined according to the EWGSOP2 criteria (Cruz-Jentoft et al., 2019). Muscle strength is considered low when handgrip strength is low (<27 kg for men or <16 kg for women) or chair stand test time is >15 s (Cruz-Jentoft et al., 2019). The presence of this criterion alone categorizes a subject as having probable sarcopenia (Cruz-Jentoft et al., 2019). When, on top of low muscle strength, muscle mass is low (skeletal muscle mass index or SMI <7.0 kg/m^2 for men or <5.5 kg/m^2 for women), the subject has confirmed sarcopenia. When gait speed is also low (<0.8 m/s), sarcopenia is considered severe. Subjects with probable, confirmed, or severe sarcopenia were included in this analysis.

2.4. Inflammatory markers

Fasted blood samples were drawn at baseline visit. Samples were centrifuged as soon as possible after venepuncture at 1200*g* for 10 min (Temp 24 °C) to collect serum samples, were transferred into cryotubes of 2 mL (Sarstedt SC Micro Tube Protein LB) and preserved at −80 °C until batch analysis. All samples were analysed for IL-1β, IL-6, IL-8 and TNF-α according to the protocol of the manufacturer in duplicate by using a validated multiplex assay (Meso Scale Diagnostics Inc., Maryland, USA, Cat. # K15053G). Three quality controls were analysed each run with between brackets the reference values for control 1, 2 & 3 (IL-1β: 218, 53 & 11.2 pg/mL, IL-6: 258, 47.7 & 7.49 pg/mL, IL-8: 239, 38.1 & 6.47 pg/mL and TNF-α: 83, 15.4 & 1.88 pg/mL). The intra-assay and inter-assay variability of the three quality controls were below 6 % and 8 %, respectively.

Plasma CRP and albumin in plasma were measured using Roche Cobas c702 (Roche Diagnostics, Basel, Switzerland) according to the manufacturer's instructions in the hospital laboratories of UZ Leuven, Belgium.

2.5. Physical activity and quality of life (QoL)

Physical activity was measured through the Dynaport MoveMonitor+ (McRoberts, The Netherlands), with build-in tri-axial accelerometer. This technology is validated for use in community-dwelling older adults (Dijkstra et al., 2010). Subjects were instructed to wear this device on five consecutive days prior to baseline visit. In case that the device was worn fewer days, measurement was excluded from the analysis. Total steps per day were calculated using the device software of the manufacturer.

To evaluate quality of life, two instruments were used, namely the Short Form - 36 Health Survey (SF-36) and the Sarcopenia Quality of Life questionnaire (SarQoL). SF-36 is a generic health-related QoL instrument that includes eight health domains: physical functioning, role limitations due to physical and emotional health, mental health, bodily pain, general health, vitality and social functioning (Ware and Sherbourne, 1992). These items make use of a norm-based scoring method, combining these eight health domains into component summary scales scores for mental and physical QoL, with higher scores suggesting better QoL. Summary scores were calculated using the STATA module “sf36”, as described elsewhere (Ryan, 1999). SarQoL is a specific health-related QoL questionnaire designed and validated for use in older adults with sarcopenia, assessing seven health domains: physical and mental health, locomotion, body composition, functionality, activities of daily living, leisure activities and fears. These domains are combined into a total SarQoL score, ranging from 0 to 100 with a higher score indicating better QoL (Beaudart et al., 2017).

2.6. Statistics

To summarize subjects' baseline characteristics descriptive statistics were used. Normality was determined through the Shapiro-Wilk test. Differences in levels of inflammatory markers according to gender and sarcopenia stages (probable vs. confirmed + severe sarcopenia) were evaluated through a Mann-Whitney *U* test, with a *p*-value <0.05 considered significant. Coefficient of variation (CV) was calculated as standard deviation / mean * 100. Pairwise calculation of the Spearman's rank correlation (ρ) was used to examine correlations. Subgroup analyses according to gender were performed. Due to the exploratory nature of the study, no correction for multiple testing was applied. A correlation was considered significant if p-value <0.05. The correlation coefficient (ρ) was interpreted as poor if <0.3, fair if ≥0.3 and <0.6, moderate if ≥0.6 and <0.8 and very strong if ≥0.8 (Akoglu, 2018). Results were presented in a heat plot. Statistics were performed using STATA SE 16.1.

3. Results

3.1. Subjects

In total, 40 older adults (15 men and 25 women) with probable, confirmed, or severe sarcopenia were included. An overview of baseline characteristics can be found in Table 1. The participants had a mean age of 77.1 ± 6.8 years (range: 65–96 years). Out of 40 subjects, 18 had probable sarcopenia, 19 confirmed and three severe sarcopenia. In seven subjects, data on physical activity were missing, whereas two and six participants had missing data of the SF-36 questionnaire and SarQoL, respectively. Participants with missing data were excluded from the analysis of which they missed data.

3.2. Inflammatory markers

Details on the levels of inflammatory markers are presented in Table 2. For three IL-6, four IL-1β, ten TNF-α and none of the IL-8 samples levels were below the detection range and were omitted from the respective results. The CV was high (range 92.54 %-197.54 %), except for albumin (6.92 %). A Mann-Whitney *U* test demonstrated no significant differences in levels of inflammatory markers according to gender. In Table 3 an overview is provided of the inflammatory according to sarcopenia stages. Subjects with probable sarcopenia had significantly ($p = 0.042$) higher levels of IL-1β (0.40 [0.11–1.87] pg/mL) than those with confirmed or severe sarcopenia (0.10 [0.06–0.21] pg/mL).

Table 1
Baseline characteristics.

Variable	All participants Mean (SD)	Women Mean (SD)	Men Mean (SD)
Age (years)	77.1 (6.8)	76.7 (5.8)	77.8 (8.5)
Number of male/female subject	40	25	15
Weight (kg)	71.7 (16.5)	69.1 (15.4)	76.1 (17.8)
Body mass index (kg/m^2)	25.9 (4.9)	26.3 (5.4)	25.1 (4.0)
Chair stand test time (s)	19.9 (5.7)	20.3 (5.1)	19.2 (6.8)
Handgrip strength (kg)	24.9 (8.8)	22.8 (7.2)	28.4 (10.3)
Appendicular lean mass [aLM] (kg)	16.86 (4.43)	14.7 (2.5)	20.5 (4.6)
Gait speed (m/s)	0.98 (0.18)	1.00 (0.18)	0.95 (0.17)
Short Physical Performance Battery score [SPPB]	8.30 (1.88)	8.08 (1.71)	8.67 (2.16)
Mean number of steps/day	6177 (5088)	6244 (4827)	6058 (5737)
SF-36 Physical component summary score	38.93 (9.12)	37.94 (8.51)	40.63 (10.18)
SF-36 Mental component summary score	51.00 (10.32)	51.19 (9.73)	50.67 (11.63)
SarQoL total score	59.42 (14.26)	55.38 (11.96)	65.93 (15.69)
Sarcopenia status (n)			
Probable sarcopenia	18 (45.0 %)	12 (48.0 %)	6 (40.0 %)
Confirmed sarcopenia	19 (47.5 %)	11 (44.0 %)	8 (53.3 %)
Severe sarcopenia	3 (7.5 %)	2 (8.0 %)	1 (6.7 %)

Table 2
Levels of inflammatory markers in the participants.

Inflammatory marker	All participants	Women	Men	p-Value[a]	CV	N	Laboratory reference value
	Median [IQR]	Median [IQR]	Median [IQR]		(%)		
	$n = 40$	$n = 25$	$n = 15$				
CRP (mg/L)	1.00 [0.65–1.30]	1.10 [0.60–1.30]	1.00 [0.80–1.20]	0.966	92.54	40	≤5 mg/L
Albumin (g/L)	43.65 [42.10–46.20]	44.90 [42.00–46.10]	43.60 [42.20–46.40]	0.911	6.92	40	[35.0–52.0 g/L]
IL-1β (pg/mL)	0.12 [0.07–0.73]	0.15 [0.07–1.87]	0.11 [0.07–0.47]	0.591	142.17	36	∞
IL-6 (pg/mL)	1.09 [0.76–1.60]	0.90 [0.57–1.60]	1.17 [1.04–1.83]	0.159	99.69	37	∞
IL-8 (pg/mL)	14.09 [7.40–20.10]	13.06 [6.80–19.09]	14.78 [7.50–21.12]	0.567	197.54	40	∞
TNF-α (pg/mL)	2.07 [1.79–3.63]	2.09 [1.91–3.63]	1.98 [1.72–5.28]	0.325	195.60	33	∞

CRP = C-reactive protein; IL = Interleukin; TNF-α = Tumour Necrosis Factor α; IQR = Interquartile range; CV = coefficient of variation; ∞ no reference available for this population.
*p < 0.05.
[a] Mann-Whitney *U* test Women vs. Men.

Table 3
Levels of inflammatory markers in participants according to sarcopenia stages.

Inflammatory marker	All participants	Probable sarcopenia	Confirmed & severe sarcopenia	P-value[a]
	Median [IQR]	Median [IQR]	Median [IQR]	
	N = 40	N = 18	N = 22	
CRP (mg/L)	1.00 [0.65–1.30]	1.00 [0.70–1.30]	1.00 [0.60–1.30]	0.712
Albumin (g/L)	43.65 [42.10–46.20]	43.20 [42.20–46.10]	45.10 [42.00–46.20]	0.673
IL-1β (pg/mL)	0.12 [0.07–0.73]	0.40 [0.11–1.87]	0.10 [0.06–0.21]	0.042*
IL-6 (pg/mL)	1.09 [0.76–1.60]	1.16 [0.82–3.78]	1.09 [0.68–1.55]	0.327
IL-8 (pg/mL)	14.09 [7.40–20.10]	15.40 [5.53–97.37]	14.09 [9.59–16.80]	0.568
TNF-α (pg/mL)	2.07 [1.79–3.63]	2.20 [1.83–3.63]	2.00 [1.74–4.16]	0.382

CRP = C-reactive protein; IL = Interleukin; TNF-α = Tumour Necrosis Factor α; IQR = Interquartile range.
[a] Mann-Whitney *U* test Probable sarcopenia vs. confirmed and severe sarcopenia.
* $p < 0.05$.

3.3. Sarcopenia-defining parameters

Correlations of inflammatory markers and sarcopenia-related traits are presented in Fig. 1. Regarding the sarcopenia-defining parameters, IL-1β was found to have a fair, positive correlation with handgrip strength (ρ: 0.376; $p = 0.024$). Thus, suggesting that with increasing levels of IL-1β, handgrip strength was higher in these older adults with sarcopenia. Similarly, IL-6 was fairly and positively correlated with aLM (ρ: 0.334; $p = 0.0433$). No other significant correlations with the remainder of sarcopenia-defining parameters (chair stand test, gait speed or SPPB) were found.

In order to explore gender differences, calculation of Spearman's correlation coefficients was repeated in subgroups by gender, presented in Fig. 2, panel A for women and Fig. 2, panel B for men. This demonstrated that the aforementioned correlation between IL-1β and handgrip strength was significant and moderate in women (ρ: 0.629; $p = 0.001$) but not in men (ρ: 0.049; $p = 0.880$). Additionally, IL-1β correlated positively with aLM in women (ρ: 0.517; $p < 0.01$) but not in men (ρ: 0.035; $p = 0.914$). Whereas IL-8 did not have a significant correlation when examining the whole study population, subgroup analyses demonstrated a significant fair, inverse correlation with grip strength (ρ: -0.425; $p = 0.034$) in women that was opposite to the non-significant positive correlation found in men (ρ: 0.325; $p = 0.237$). The association of IL-6 with aLM in the whole study cohort failed to reach the

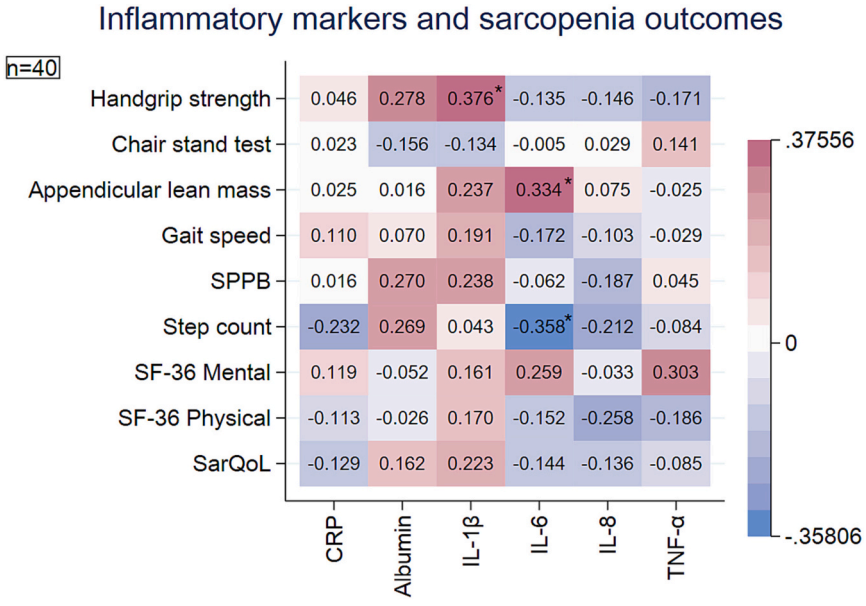

Fig. 1. Heat plot representing correlation coefficients (ρ) between sarcopenia-related traits and inflammatory markers in all participants
* = p < 0.05; SPPB = Short Physical Performance Battery; CRP = C-reactive protein; IL = Interleukin; TNF-α = Tumour Necrosis Factor α.

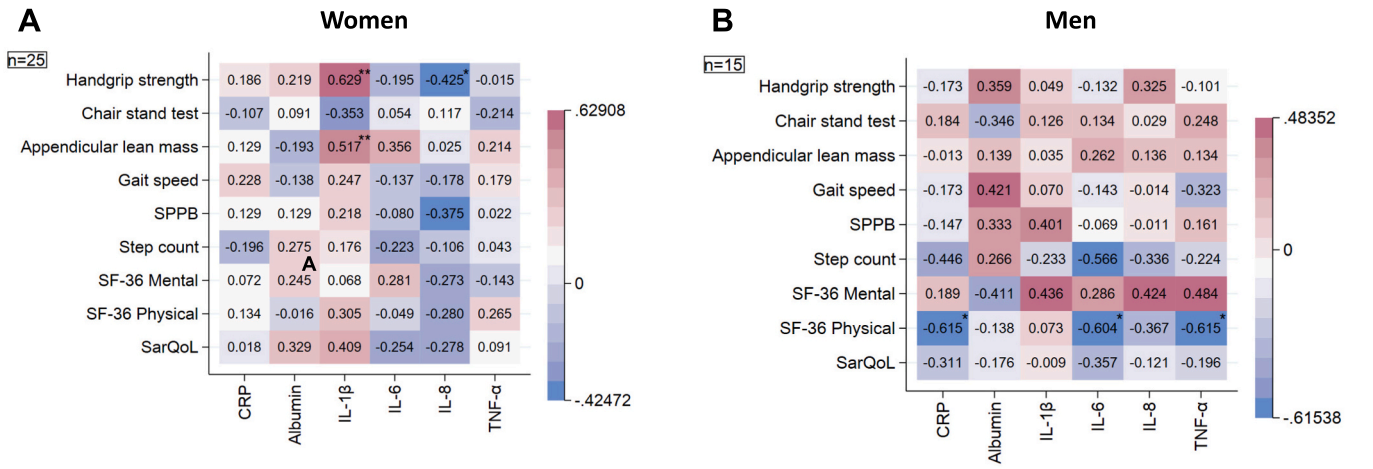

Fig. 2. Heat plot representing correlation coefficients (ρ) between sarcopenia-related traits and inflammatory according to gender.
* = p < 0.05; ** = p < 0.01; SPPB = Short Physical Performance Battery; CRP = C-reactive protein; IL = Interleukin; TNF-α = Tumour Necrosis Factor α.

significance threshold when examined separately according to gender (ρ: 0.356; $p = 0.096$ for women and ρ: 0.262; $p = 0.367$ for men).

3.4. Physical activity

The participants had a mean step count of 6177 (±5088), although the large standard deviation suggests a large interpersonal variability. When observing correlations between step count and inflammatory markers, a significant and fair but inverse correlation with IL-6 was found (ρ: -0.358; $p = 0.048$). However, when analysing according to gender, this correlation lost significance (ρ: -0.223; $p = 0.359$ for women and ρ: -0.566; $p = 0.055$ for men). No significant correlations of step count with the other inflammatory markers (CRP, albumin, IL-1β, IL-8 or TNF-α) were found.

3.5. Quality of life

The results of the SF-36 questionnaires were summarized into standardised component scores, both a physical and a mental one (Table 1). When examining both genders together, no significant associations with QoL outcomes could be found. In men, however, moderate and significant but inverse correlations were found between the pro-inflammatory cytokines CRP (ρ: -0.615; $p = 0.019$), IL-6 (ρ: -0.604; $p = 0.029$), TNF-α (ρ: -0.615; $p = 0.025$) and the SF-36 physical component scores. Furthermore, no significant associations with the SF-36 mental component scores, nor with the more sarcopenia-specific SarQoL questionnaire scores were present.

4. Discussion

To the best of our knowledge, this study was the first to examine correlations between inflammatory markers and various sarcopenia-related traits in a population of sarcopenic older adults, diagnosed according to the EWGSOP2. Contrary to what was expected, our findings suggest a positive correlation of IL-1β levels with handgrip strength and of IL-6 levels with aLM, instead of an inverse relationship. On the other hand, step count was – as expected – inversely related with IL-6 levels. Furthermore, our data highlights a potential role of gender, as the gender-specific analyses in the present study revealed altered results compared to the whole study population. In women, IL-1β was positively correlated with handgrip strength and appendicular lean mass, whereas IL-8 was inversely correlated with handgrip strength. In the men included in this study, no significant associations were found with sarcopenia-defining parameters (handgrip strength, chair stand test, aLM, gait speed and SPPB) but only an inverse correlation of pro-inflammatory markers CRP, IL-6 and TNF-α with the SF-36 physical component score, representing physical-related QoL.

The level of the inflammatory markers examined in the present study suggested an absence of acute high inflammation, with median CRP 1.00 mg/L and IQR [0.65–1.30] mg/L [reference of physiological levels ≤5 mg/L]. These values are compatible with the previously reported range of plasma CRP that characterizes Inflammageing in older adults (>70 years), which is lower than in case of an acute infection but higher compared to younger adults (20–49 years) (Alberro et al., 2021). Head-to-head comparison of the levels of the inflammatory cytokines of the sarcopenic older adults (according to EWGSOP2) in our study with previous data is hard, since previous studies vary in assays used to determine inflammatory cytokines and the populations they examined. Nonetheless, when we compare the values of sarcopenic older adults from ENHANce (EWGSOP2) with those of sarcopenic patients (defined as having low muscle mass) with chronic obstructive pulmonary disease (COPD) (Joppa et al., 2016), the median levels of IL-6 (1.09 [0.76–1.60] versus 1.3 [0.7–2.6] pg/mL) and TNF-α (2.07 [1.79–3.63] versus 2.4 [2.4–21.9] pg/mL) are in similar range. In contrast, the median IL-8 levels are higher in ENHANce participants compared to the COPD patients (14.09 [7.40–20.10] versus 7.5 [3.6–14.1] pg/mL) (Joppa et al., 2016). Of all the examined inflammatory markers, albumin was the only inflammatory marker that did not demonstrate a significant association with an examined sarcopenia-related trait. Albumin can be considered an inflammatory marker due to its properties as a negative acute phase protein but is also a biomarker of nutritional status (Cabrerizo et al., 2015; Cheong et al., 2020). All participants had albumin levels within laboratory reference of normal values [35.0–52.0 g/L]. This, together with being a non-specific inflammatory marker, might explain the lack of findings with albumin in the present study.

Gender plays an important role in the results found in the present study. Although we did not find a significant gender difference in the levels of inflammatory markers in the included participants, gender altered the correlations found between the examined inflammatory markers and sarcopenia-related traits. This is in line with previous research suggesting that gender-specific inflammatory profiles may exist when looking into the presence of sarcopenia. The previously mentioned study of Marzetti et al. found gender-specific "profiles" for PF&S. More specifically, in women, PF&S is more associated with higher levels CRP, P-selectin and MIP-1β combined with lower levels of MPO, MCP-1, PDGF, MIP-1α and IL-8. In contrast, men with PF&S were characterized by increased levels of CRP and decreased levels of interferon-γ, fibroblast growth factor basic, IL-17, MIP-1β and TNF-α (Marzetti et al., 2019). In the same line of thought, the magnitude of the association between biomarkers (e.g., vitamin D) and sarcopenia has been suggested

to differ between men and women (Petermann-Rocha et al., 2020; Berghella et al., 2014). For example, a recent meta-analysis examining the correlation and gender differences of plasma IL-6 with muscle strength in healthy older adults (>65 years) demonstrated that higher cut-offs for IL-6 levels in men might be needed – compared to women – in order to predict adverse outcomes (e.g., low handgrip strength) or sarcopenia (Mikó et al., 2018). In the whole ENHANce cohort, IL-6 levels were (significantly) positively correlated with aLM. When examining this association according to gender, it was stronger in women than men (ρ: 0.356 for women compared to ρ: 0.262 for men), although both losing significance in the subgroup analysis. IL-8 levels were inversely associated with handgrip strength in women, but not in men. Thus, suggesting that with increasing levels of IL-8, female participants had lower grip strength. This fits in the knowledge that the pro-inflammatory cytokine IL-8 is associated with a higher risk on sarcopenia in community-dwelling older adults (age 50–70 years) (Westbury et al., 2018), but contradicts with the findings of Marzetti et al. that PF&S is characterized by lower levels of IL-8 in women (Marzetti et al., 2019). Interestingly, the women in our study demonstrated significant correlations of inflammatory markers with 'firm' sarcopenia-defining parameters like handgrip strength and aLM, whereas the men exerted correlations with more 'secondary' sarcopenia-related traits such as the SF-36 physical QoL score. The gender differences in the demonstrated correlations might be partly attributed to the small sample size with relatively smaller number of men included in present study, but still deserve further investigation in large-scale prospective research.

Handgrip strength – one of the principal *sarcopenia-defining parameters* – was positively associated with IL-1β (whole cohort & women) and inversely associated with IL-8 (women only). Since IL-1β and IL-8 are both pro-inflammatory cytokines, the positive direction of the IL-1β correlation is surprising. Similarly, IL-1β was positively associated with aLM in the ENHANce participants. This fits with IL-1β levels being higher in participants with probable sarcopenia (normal muscle mass) compared to those with confirmed or severe sarcopenia (who have lower muscle mass). However, one would expect an inverse association of IL-1β with these parameters due to its pro-inflammatory properties since IL-1β has been known to block differentiation of human myoblasts into myotubes (Trendelenburg et al., 2012) and as such one should expect it to play a negative role in aLM. In addition, a lower expression of interleukin 1 receptor (IL1R) – the main receptor for IL-1β – in skeletal muscle has been described to be associated with higher grip strength in older men from the Hertfordshire study (Patel et al., 2014). However, a recent study examining the association of IL-1β with SMI and grip strength in community-dwelling older adults could not confirm any association with these parameters (Liu et al., 2021). Similarly contradictious as IL-1β, the IL-6 levels also demonstrated a positive association with aLM in our community-dwelling sarcopenic older adults of the ENHANce study. This in sharp contrast with previous data suggesting an inverse relationship with lean mass in mobility-limited older adults (Grosicki et al., 2020), older adults with a high cardiovascular risk profile (Cesari et al., 2005) or well-functioning older adults (Visser et al., 2002). These contradicting results might be – at least partly – attributed to the population in which the association was explored. For example, a recent meta-analysis exploring biomarkers for frailty found that IL-6 levels were higher in frail community-dwelling and hospitalized persons compared to their robust counterparts, whereas IL-6 levels were lower in frail institutionalised older adults compared to the robust peers (Picca et al., 2022). Furthermore – as previously mentioned – the two-headed action of IL-6 can play a role in the contradicting results, since IL-6 exerts different effects (pro-inflammatory or anti-inflammatory) depending on the levels and duration of exposure (Belizário et al., 2016; Calabrese and Rose-John, 2014). To illustrate, chronically increased IL-6 levels are known to promote skeletal muscle wasting (Belizário et al., 2016). However, this is in contrast with our findings of higher IL-6 levels with increasing appendicular lean mass. This might be explained by the fact that IL-6 can also be considered a myokine – secreted by muscle tissue (Karstoft and Pedersen, 2016) – and as such with increasing amount of muscle mass, more IL-6 might be secreted. Longitudinal research of this association in a sarcopenic population is needed to elucidate this issue.

Physical activity expressed as step count was inversely associated with IL-6 levels in ENHANce participants, suggesting lower mean step count of our participants with increasing IL-6. Whereas exercise can induce an acute IL-6 increase (MacNeil et al., 2021), promoting physical activity can lower IL-6 levels on the long run, as recently demonstrated in persons with prediabetes (Jadhav et al., 2021). This association highlights the importance of recommending regular physical activity (including aerobic exercise such as walking) to older adults, as stated in the recent ICFSR exercise recommendations (Izquierdo et al., 2021). In general, our participants had a rather low mean daily step count (6177 ± 5088 steps/day), in contrast to the recommendation that healthy older adults should walk at least 7000 to 10,000 steps per day (Tudor-Locke et al., 2011). In view of the abovementioned gender differences found in the present study, we explored whether step count was different according to gender but found no significant gender difference in physical activity (data not shown).

Quality of life related to physical function – expressed through the SF-36 physical component score – was inversely correlated with pro-inflammatory cytokines CRP, IL-6 and TNF-α in the sarcopenic men of present study, but not in the women. It is unclear why this association is present in men but not women. In women, this association is not only insignificant but also inverse (for CRP and TNF-α). A recent study of Fábrega-Cuadros et al. found that community-dwelling middle-aged and older women (>50y) scored better in the SF-36 domains physical functioning, physical role, and mental health (Fábrega-Cuadros et al., 2021). However, in present study no significant gender-differences in SF-36 scores could be found (data not shown). Nonetheless, our findings match with recent data from the EMAS study confirming an inverse association of pro-inflammatory CRP and white blood cell count with the SF-36 physical component score in middle-aged and older men (Dupont et al., 2021). Similarly, lower scores of the RAND questionnaire – strongly related to the SF-36 – were demonstrated to be related with higher inflammation levels (IL-6 and CRP) in healthy older adults (Christian et al., 2011). When observing the correlations with the more sarcopenia-specific SarQoL questionnaire, men demonstrated a similar trend as with the SF-36 physical component score for CRP, IL-6 and TNF-α, but it did not reach the significance level. Future research is needed to unravel these findings.

A *strength* of present study is that it is the first to examine associations of this variety of inflammatory markers with several sarcopenia-related traits in a population of older adults with sarcopenia defined using the EWGSOP2 criteria. The exploratory nature of this study will help guide further targeted research and the results stress the importance to perform gender-specific analyses. To limit the influence of preanalytical conditions on the results, a standard operating procedure (SOP) was made for sample processing of all includes samples. Moreover, the objectively measured physical activity and presence of several QoL-related traits add to the value of the study.

One of the *limitations* of this study is the small study sample, next to the cross-sectional character of the study and lack of longitudinal data. The small sample size also implies it is not excluded that the associations found are spurious. Additionally, as demonstrated through the high CV in the various inflammatory markers, the inflammatory profiles of the participants were quite divers despite being a uniformly well-defined study population. Moreover, the inclusion of less men than women in as well as the small sample size might have underpowered the study, thus making it hard to drawn firm conclusions regarding the relationship of inflammatory markers with sarcopenia-related traits. Finally, despite examining no less than six inflammatory markers, various other cytokines (e.g., IL-4, IL-10, IL-13…) might also play a role in sarcopenia and its related traits and deserve exploration in future studies.

5. Conclusion

This exploratory study demonstrated subclinical low levels of inflammatory markers in older adults suffering from sarcopenia, compatible with age-related Inflammageing. Some interesting associations (expected and not expected) were found between the examined inflammatory markers and sarcopenia-related traits. Although no gender-differences in levels of inflammation were found, gender had a significant influence on the associations between these inflammatory markers and sarcopenia-related traits. The findings of the present study stress the complexity of the inflammageing-sarcopenia interplay and the importance of not only looking at muscle mass or the sarcopenia construct when researching sarcopenia, but also considering other sarcopenia-related traits and gender in future research.

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Ethics approval

The study was approved by KU Leuven/UZ Leuven Ethics Committee (s60763). All participants provided written informed consent.

CRediT authorship contribution statement

Jolan Dupont: Conceptualization, Data curation, Formal analysis, Writing – original draft, Writing – review & editing. **Laura Vercauteren:** Writing – review & editing. **Nadjia Amini:** Writing – review & editing. **Laurence Lapauw:** Writing – review & editing. **Maxim De Schaepdryver:** Data curation, Formal analysis, Writing – review & editing. **Koen Poesen:** Writing – review & editing. **Lenore Dedeyne:** Data curation, Writing – review & editing. **Sabine Verschueren:** Writing – review & editing. **Jos Tournoy:** Writing – review & editing. **Katrien Koppo:** Writing – review & editing. **Evelien Gielen:** Conceptualization, Writing – review & editing.

Declaration of competing interest

EG is a board member of the Belgian Bone Club. EG has received consultancy fees and lectures fees from Alexion, Amgen, Sandoz, Takeda, UCB, unrelated to this work. None of the other authors have any conflicts of interest to declare related to this manuscript.

Data availability

The data that has been used is confidential.

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References

Akoglu, H., 2018. User's guide to correlation coefficients. Turk. J. Emerg. Med. 18 (3), 91–93.
Alberro, A., Iribarren-Lopez, A., Sáenz-Cuesta, M., Matheu, A., Vergara, I., Otaegui, D., 2021. Inflammaging markers characteristic of advanced age show similar levels with frailty and dependency. Sci. Rep. 11 (1), 4358.
Bano, G., Trevisan, C., Carraro, S., Solmi, M., Luchini, C., Stubbs, B., et al., 2017. Inflammation and sarcopenia: a systematic review and meta-analysis. Maturitas 96, 10–15.
BCGuidelines.ca, 2017. Frailty in Older Adults – Early Identification and Management.
Beaudart, C., Biver, E., Reginster, J.Y., Rizzoli, R., Rolland, Y., Bautmans, I., et al., 2017. Validation of the SarQoL(R), a specific health-related quality of life questionnaire for sarcopenia. J. Cachexia. Sarcopenia Muscle 8 (2), 238–244.
Belizário, J.E., Fontes-Oliveira, C.C., Borges, J.P., Kashiabara, J.A., Vannier, E., 2016. Skeletal muscle wasting and renewal: a pivotal role of myokine IL-6. Springerplus 5, 619.
Berghella, A.M., Contasta, I., Marulli, G., D'Innocenzo, C., Garofalo, F., Gizzi, F., et al., 2014. Ageing gender-specific "Biomarkers of Homeostasis", to protect ourselves against the diseases of the old age. Immun. Ageing 11 (1), 3.
Cabrerizo, S., Cuadras, D., Gomez-Busto, F., Artaza-Artabe, I., Marín-Ciancas, F., Malafarina, V., 2015. Serum albumin and health in older people: review and meta analysis. Maturitas 81 (1), 17–27.
Calabrese, L.H., Rose-John, S., 2014. IL-6 biology: implications for clinical targeting in rheumatic disease. Nat. Rev. Rheumatol. 10 (12), 720–727.
Cesari, M., Kritchevsky, S.B., Baumgartner, R.N., Atkinson, H.H., Penninx, B.W., Lenchik, L., et al., 2005. Sarcopenia, obesity, and inflammation–results from the trial of angiotensin converting enzyme inhibition and novel cardiovascular risk factors study. Am. J. Clin. Nutr. 82 (2), 428–434.
Cheong, M., Chew, S.T.H., Oliver, J., Baggs, G., Low, Y.L., How, C.H., et al., 2020. Nutritional biomarkers and associated factors in community-dwelling older adults: findings from the SHIELD study. Nutrients 12 (11).
Christian, L.M., Glaser, R., Porter, K., Malarkey, W.B., Beversdorf, D., Kiecolt-Glaser, J.K., 2011. Poorer self-rated health is associated with elevated inflammatory markers among older adults. Psychoneuroendocrinology 36 (10), 1495–1504.
Cruz-Jentoft, A.J., Bahat, G., Bauer, J., Boirie, Y., Bruyere, O., Cederholm, T., et al., 2019. Sarcopenia: revised European consensus on definition and diagnosis. Age Ageing 48 (1), 16–31.
Dalle, S., Rossmeislova, L., Koppo, K., 2017. The role of inflammation in age-related sarcopenia. Front. Physiol. 8, 1045.
Dedeyne, L., Dupont, J., Koppo, K., Verschueren, S., Tournoy, J., Gielen, E., 2020. Exercise and nutrition for healthy AgeiNg (ENHANce) project - effects and mechanisms of action of combined anabolic interventions to improve physical functioning in sarcopenic older adults: study protocol of a triple blinded, randomized controlled trial. BMC Geriatr. 20 (1), 532.
Dijkstra, B., Kamsma, Y., Zijlstra, W., 2010. Detection of gait and postures using a miniaturised triaxial accelerometer-based system: accuracy in community-dwelling older adults. Age Ageing 39 (2), 259–262.
Dupont, J., Antonio, L., Dedeyne, L., O'Neill, T.W., Vanderschueren, D., Rastrelli, G., et al., 2021. Inflammatory markers are associated with quality of life, physical activity, and gait speed but not sarcopenia in aged men (40–79 years). J. Cachexia. Sarcopenia Muscle 12 (6), 1818–1831.
von Elm, E., Altman, D.G., Egger, M., Pocock, S.J., Gøtzsche, P.C., Vandenbroucke, J.P., 2007. The strengthening the reporting of observational studies in epidemiology (STROBE) statement: guidelines for reporting observational studies. PLoS Med. 4 (10), e296.
Fábrega-Cuadros, R., Hita-Contreras, F., Martínez-Amat, A., Jiménez-García, J.D., Achalandabaso-Ochoa, A., Lavilla-Lerma, L., et al., 2021. Associations between the severity of sarcopenia and health-related quality of life in community-dwelling middle-aged and older adults. Int. J. Environ. Res. Public Health 18 (15).
Folstein, M.F., Folstein, S.E., McHugh, P.R., 1975. "Mini-mental state". A practical method for grading the cognitive state of patients for the clinician. J. Psychiatr. Res. 12 (3), 189–198.
Franceschi, C., Bonafe, M., Valensin, S., Olivieri, F., De Luca, M., Ottaviani, E., et al., 2000. Inflammaging. An evolutionary perspective on immunosenescence. Ann. N. Y. Acad. Sci. 908, 244–254.
Grosicki, G.J., Barrett, B.B., Englund, D.A., Liu, C., Travison, T.G., Cederholm, T., et al., 2020. Circulating Interleukin-6 is associated with skeletal muscle strength, quality, and functional adaptation with exercise training in mobility-limited older adults. J. Frailty Aging 9 (1), 57–63.
Guralnik, J.M., Simonsick, E.M., Ferrucci, L., Glynn, R.J., Berkman, L.F., Blazer, D.G., et al., 1994. A short physical performance battery assessing lower extremity function: association with self-reported disability and prediction of mortality and nursing home admission. J. Gerontol. 49 (2), M85–M94.
Izquierdo, M., Merchant, R.A., Morley, J.E., Anker, S.D., Aprahamian, I., Arai, H., et al., 2021. International exercise recommendations in older adults (ICFSR): expert consensus guidelines. J. Nutr. Health Aging 25 (7), 824–853.
Jadhav, R.A., Maiya, G.A., Hombali, A., Umakanth, S., Shivashankar, K.N., 2021. Effect of physical activity promotion on adiponectin, leptin and other inflammatory markers in prediabetes: a systematic review and meta-analysis of randomized controlled trials. Acta Diabetol. 58 (4), 419–429.
Joppa, P., Tkacova, R., Franssen, F.M., Hanson, C., Rennard, S.I., Silverman, E.K., et al., 2016. Sarcopenic obesity, functional outcomes, and systemic inflammation in patients with chronic obstructive pulmonary disease. J. Am. Med. Dir. Assoc. 17 (8), 712–718.
Karstoft, K., Pedersen, B.K., 2016. Skeletal muscle as a gene regulatory endocrine organ. Curr. Opin. Clin. Nutr. Metab. Care 19 (4), 270–275.
Lampignano, L., Bortone, I., Castellana, F., Donghia, R., Guerra, V., Zupo, R., et al., 2021. Impact of different operational definitions of sarcopenia on prevalence in a

population-based sample: the salus in Apulia study. Int. J. Environ. Res. Public Health 18 (24).
Liu, H.C., Han, D.S., Hsu, C.C., Wang, J.S., 2021. Circulating MicroRNA-486 and MicroRNA-146a serve as potential biomarkers of sarcopenia in the older adults. BMC Geriatr. 21 (1), 86.
MacNeil, L.G., Tarnopolsky, M.A., Crane, J.D., 2021. Acute, exercise-induced alterations in cytokines and chemokines in the blood distinguish physically active and sedentary aging. J. Gerontol. A Biol. Sci. Med. Sci. 76 (5), 811–818.
Marzetti, E., Picca, A., Marini, F., Biancolillo, A., Coelho-Junior, H.J., Gervasoni, J., et al., 2019. Inflammatory signatures in older persons with physical frailty and sarcopenia: the frailty "cytokinome" at its core. Exp. Gerontol. 122, 129–138.
Mikó, A., Pótó, L., Mátrai, P., Hegyi, P., Füredi, N., Garami, A., et al., 2018. Gender difference in the effects of interleukin-6 on grip strength - a systematic review and meta-analysis. BMC Geriatr. 18 (1), 107.
Pan, L., Xie, W., Fu, X., Lu, W., Jin, H., Lai, J., et al., 2021. Inflammation and sarcopenia: a focus on circulating inflammatory cytokines. Exp. Gerontol. 154, 111544.
Patel, H.P., Al-Shanti, N., Davies, L.C., Barton, S.J., Grounds, M.D., Tellam, R.L., et al., 2014. Lean mass, muscle strength and gene expression in community dwelling older men: findings from the hertfordshire sarcopenia study (HSS). Calcif. Tissue Int. 95 (4), 308–316.
Pedersen, B.K., Fischer, C.P., 2007. Physiological roles of muscle-derived interleukin-6 in response to exercise. Curr. Opin. Clin. Nutr. Metab. Care 10 (3), 265–271.
Petermann-Rocha, F., Gray, S.R., Pell, J.P., Celis-Morales, C., Ho, F.K., 2020. Biomarkers profile of people with sarcopenia: a cross-sectional analysis from UK biobank. J. Am. Med. Dir. Assoc. 21(12):2017.e1-.e9.
Picca, A., Coelho-Junior, H.J., Calvani, R., Marzetti, E., Vetrano, D.L., 2022. Biomarkers shared by frailty and sarcopenia in older adults: a systematic review and meta-analysis. Ageing Res. Rev. 73, 101530.
Roberts, H.C., Denison, H.J., Martin, H.J., Patel, H.P., Syddall, H., Cooper, C., et al., 2011. A review of the measurement of grip strength in clinical and epidemiological studies: towards a standardised approach. Age Ageing 40 (4), 423–429.
Ryan, P., 1999. SF36: Stata module to calculate summary statistics for the SF-36 Health Survey Instrument.
Schaap, L.A., Pluijm, S.M., Deeg, D.J., Visser, M., 2006. Inflammatory markers and loss of muscle mass (sarcopenia) and strength. Am. J. Med. 119 (6), 526.e9-17.
Schaap, L.A., Pluijm, S.M., Deeg, D.J., Harris, T.B., Kritchevsky, S.B., Newman, A.B., et al., 2009. Higher inflammatory marker levels in older persons: associations with 5-year change in muscle mass and muscle strength. J. Gerontol. A Biol. Sci. Med. Sci. 64 (11), 1183–1189.
Shokri-Mashhadi, N., Moradi, S., Heidari, Z., Saadat, S., 2021. Association of circulating C-reactive protein and high-sensitivity C-reactive protein with components of sarcopenia: a systematic review and meta-analysis of observational studies. Exp. Gerontol. 150, 111330.
Trendelenburg, A.U., Meyer, A., Jacobi, C., Feige, J.N., Glass, D.J., 2012. TAK-1/p38/nNFκB signaling inhibits myoblast differentiation by increasing levels of activin A. Skelet. Muscle 2 (1), 3.
Tudor-Locke, C., Craig, C.L., Aoyagi, Y., Bell, R.C., Croteau, K.A., De Bourdeaudhuij, I., et al., 2011. How many steps/day are enough? For older adults and special populations. Int. J. Behav. Nutr. Phys. Act. 8, 80.
Visser, M., Pahor, M., Taaffe, D.R., Goodpaster, B.H., Simonsick, E.M., Newman, A.B., et al., 2002. Relationship of interleukin-6 and tumor necrosis factor-alpha with muscle mass and muscle strength in elderly men and women: the health ABC study. J. Gerontol. A Biol. Sci. Med. Sci. 57 (5), M326–M332.
Wang, Y., Welc, S.S., Wehling-Henricks, M., Tidball, J.G., 2018. Myeloid cell-derived tumor necrosis factor-alpha promotes sarcopenia and regulates muscle cell fusion with aging muscle fibers. Aging Cell 17 (6), e12828.
Ware Jr., J.E., Sherbourne, C.D., 1992. The MOS 36-item short-form health survey (SF-36). I. Conceptual framework and item selection. Med. Care 30 (6), 473–483.
Westbury, L.D., Fuggle, N.R., Syddall, H.E., Duggal, N.A., Shaw, S.C., Maslin, K., et al., 2018. Relationships between markers of inflammation and muscle mass, strength and function: findings from the hertfordshire cohort study. Calcif. Tissue Int. 102 (3), 287–295.