Similar body composition, muscle size and strength adaptations to resistance training in lacto-ovo-vegetarians and non-vegetarians

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ABSTRACT:

There is a popular belief that meat consumption is necessary to optimize adaptations to strength training (ST), but evidence to support this hypothesis is scarce. Therefore, this study aimed to compare ST adaptations in lacto-ovo-vegetarians (LOV) and non-vegetarians (NV) with adjusted protein intake per meal. Sixty-four LOV and NV performed 12 weeks of ST and were instructed to ingest at least 20 g of protein, in each main meal during the experimental period. Quadriceps femoris muscle thickness (QFMT), knee extension one-repetition maximum (1RM) and isometric peak torque (PT), as well as participants' body composition were assessed before and after the intervention. Dietary intake was assessed throughout the study. After 12 weeks similar increases in QFMT (LOV: 9.2 ± 5.4; NV: 5.5±8.1mm), knee extension 1RM (LOV: 24.7±11.1; NV: 21.6±9.8 kg) and PT (LOV: 29.8±33.4; NV: 17.5±19.4 N·m) and lean body mass (LOV: 1.3±0.9; NV: 1.4±1.4 kg), alongside a decrease in body fat mass (LOV: -0.5±1.6; NV -0.8±1.6 kg), were observed in both groups at the end of the training period (p<0.05). LOV had lower protein consumption than NV throughout the study (p<0.05), but participants reached intake of at least 1.2 g of protein/kg/day during the experimental period. In conclusion, LOV and NV displayed similar improvements in muscle mass, strength and in body composition after 12 weeks of ST, suggesting that meat consumption and higher protein intake in NV did not bring about further benefits to early adaptations to ST.

KEYWORDS*:* Vegetarianism; Resistance training; Hypertrophy; Muscle strength; Dietary protein; Sports nutrition

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INTRODUCTION

Strength training (ST) is widely known for bringing about increases in muscle mass, strength, and improvements in body composition in healthy and clinical populations (Yoo et al. 2018; Maestroni et al. 2020). Prolonged periods of protein surplus associated with appropriate training stimulus is thought to be necessary to optimize skeletal muscle hypertrophy (Phillips 2014; Haun et al. 2019), and ingestion of 20 g (or ~0.3g/kg) of high quality protein in a meal following ST sessions has been reported to maximize acute muscle protein synthesis in young untrained adults (Moore et al. 2009, 2015; Churchward-Venne et al. 2012; Witard et al. 2014; Stokes et al. 2018). This anabolic effect may last up to 5 h, and seems to be influenced by the amino acid composition of the meal (Areta et al. 2013).

Animal protein is an excellent source of amino acids, and it is popular belief that meat consumption is necessary to optimize strength training (ST) adaptations (Brosnan and Brosnan 2016). Well-planned vegetarian diets, however, can provide all necessary nutrients for the organism. Nevertheless, reduced protein intake is commonplace among vegetarians compared to individuals who consume meat (Craig and Mangels 2009; Clarys et al. 2014; Melina et al. 2015; Elorinne et al. 2016). Hence, one may hypothesize that ST adaptations could be hampered in those adopting a meat free or plant-based diet.

Despite increase in popularity in plant-based diets worldwide (Kamiński et al. 2020), few studies have been conducted on the impact of such diets on responses to strength training programs. To date, studies with ST have investigated only non-vegetarian older adults who stopped eating meat during a short experimental period, and existing evidence is far from conclusive with reports of detrimental (Campbell et al. 1999), benefic (Wells et al. 2003) or no effect (Haub et al. 2005) on ST adaptation with restricted meat consumption.

The rapid and short-lived dietetic modifications in previous experiments, however, make it difficult for authors to tease out the effect of diet on ST adaptations, as the influence of dietary adjustments per se become a confounding factor. For example, long-term vegetarians have been reported to have lower levels of intramuscular creatine than non-vegetarians (Burke et al. 2003), and a skeletal muscle creatine concentration has been shown to decrease over the course of 21 days following a lacto-ovo-vegetarian diet (Lukaszuk et al. 2002) , which may have influenced previous studies where participants switched to meat free diets as part of the intervention (Campbell et al. 1999; Wells et al. 2003; Haub et al. 2005).

Despite increasing global interest in vegetarianism, little is known about the influence of diets without restricted meat consumption on skeletal muscle adaptation (Craddock et al. 2016; Kamiński et al. 2020). Thus, the present study aimed to investigate muscular and body composition adaptations to 12 weeks of ST in lacto-ovo-vegetarian (LOV) and non-vegetarian (NV) adults with adjusted protein intake per meal to determine whether lacto-ovo-vegetarian diet influences ST adaptations in previously untrained individuals.

PARTICIPANTS AND METHODS

Participants

Sample size was calculated using G*Power (Faul et al. 2007) based on a difference 20 kg on the bilateral knee extension one-repetition maximum (1RM) after the ST using power of 80% and type I error probability of 5%. The calculation determined a total of 64 participants (and as such, sixty-four healthy adults (44 females, and with age ranging from 19 to 55 years) participated on the present study.

Individuals should be untrained and have adopted a LOV or NV diet for at least 6 months prior to participating in the study. Exclusion criteria included individuals who consumed protein, amino acid or thermogenic supplements; adopted vegan, lacto-vegetarian or ovo-vegetarian diets; had body mass index equal or higher than 30 kg/m²; presented any chronic disease and/or musculoskeletal disorder that could limit the participation on a ST program; and not having financial conditions to adhere to dietary modifications. We opted to exclude ovo-vegetarian and lacto-vegetarian from the study due to the amino acid profile of animal food sources being largely different in these two groups, especially concerning the amount of leucine (Burke et al. 2012). Participants were carefully informed of the purpose, procedures, and risks of the study, and a written informed consent was obtained from all participants. All procedures were approved by the local ethics committee (register number: 69787617.0.0000.5327) and conducted in accordance with the Declaration of Helsinki.

Experimental design

The present study is a 3-month non-randomized clinical trial with two arms: LOV and NV, registered at clinicaltrials.gov as NCT03785002. Both groups performed 12 weeks of a total-body ST program, 2x per week in non-consecutive days (Figure 1). Participants were required to not change their diet (i.e. changing from NV to LOV), but nutritional adjustments of protein intake in accordance with their food habits (i.e. vegetarian or non-vegetarian diet) were made in attempt to control for protein intake and minimize confounding factors. The following outcomes were measured before (PRE) and after (POST) the intervention period: quadriceps femoris muscle thickness (QFMT), maximal isometric and dynamic knee extension strength, body composition, and dietary intake. Dietary intake was also assessed two more times during the training period. All assessments were conducted at the Exercise Research Laboratory (LAPEX) of Federal University of Rio Grande do Sul (UFRGS).

Pre-training testing (PRE) was performed at baseline. Participants visited the laboratory on two occasions separated by at least 48 h. On the first visit, participants underwent assessments of body composition by dual-energy X-ray absorptiometry and ultrasonography of the quadriceps femoris. Then, participants were familiarized with testing procedures and performed the 1-repetition maximum (1RM) knee extension test. During the second visit, participants' dietary intake was assessed, their isometric knee extension strength was test tested, followed by retest of participants' knee extension 1RM. By the end of the second visit, each participant met the study nutritionists to received individual recommendations about nutritional adjustments to be taken throughout the study in order to reach the targeted protein intake per meal. Post-training (POST) testing was performed as described above, with at least 2 days of interval from the last training session. Measurements were taken by experienced investigators, which identical procedures and instructions used at PRE and POST. Excluding the nutritionist, all investigators were blinded to the participants' group. The participants were instructed not to consume any type of supplement during the study.

Strength Training Intervention

The ST program was performed twice a week for 12 weeks and all participants were advised to abstain from other forms exercises training. Both groups performed the same ST protocol, following the exercise prescription detailed in table 1. The following exercises were included in the ST routine: leg press, bench press, bilateral knee extension, biceps curl, bilateral leg curl, lat pull-down, triceps pulley, abdominal crunch, and back extension. Intensity of the ST increased every 3 weeks, with participants exercising within a target repetition maximum range and individual exercise intensity was increased whenever participants could perform more repetitions than the target range for a specific load. All sessions were supervised by fitness professionals (up to 5 participants per professional) and the volume–load (set x repetitions x load) for lower limb exercises of each participant was recorded during all sessions.

Experimental Procedures

Muscle thickness

Muscle thickness of the rectus femoris (RF), vastus intermedius (VI), vastus medialis (VM) and vastus lateralis (VL) of the right limb, were measured by ultrasonography (Nemio XG, Toshiba, Japan) using a linear-array probe with 60 mm and 9.0 MHz (image depth 70 mm, 90 dB general gain and time-gain compensation in the neutral position). The probe was coated with water-soluble transmission gel to provide acoustic contact without depressing the dermal surface. Before ultrasound imaging, participants rested in the supine position for 10 minutes with the lower limbs extended and relaxed (Lopez et al. 2019). Ultrasound image was recorded on transverse axis to the fiber direction. Muscle thickness of RF, VL and VM was defined as the distance between the subcutaneous adipose tissue-muscle interface to the deep aponeurosis of each muscle, and the VI was defined as the distance between the muscle superficial aponeurosis-bone interface. Images were analyzed using ImageJ software (National Institutes of Health, Bethesda, MD, USA). Three images of each muscle were obtained, and the average of value the three images was used for each muscle in further analysis. Quadriceps femoris muscle thickness (QF_{MT}) was calculated as the sum of the thickness of the four quadriceps femoris portions (Fukumoto et al. 2012; Pinto et al. 2014). All images were analyses by the same investigator (C.M.B), who was blinded by participant group.

Muscle Strength and Specific Tension

Bilateral knee extension 1RM was assessed using a commercial knee extension machine (KonnenGym, China) and all tests were conducted by a single investigator

(J.L.T.) Individual 1RM load was determined within five attempts. To do so, the 1RM test load was increased until a single valid repetition could no longer be performed using the correct technique. Each participant range of motion was recorded in the familiarization session with a custom-made device and the same range of motion was used for assessment throughout the study. Three to five minutes of recovery were allowed between attempts and if the 1RM test and retest differed by >5% a new testing session was performed to confirm individual 1RM (Ploutz-Snyder and Giamis 2001).

Knee extension maximal isometric peak torque (PT) was assessed in the right leg using an isokinetic dynamometer (Cybex Norm, NY, USA), calibrated according to the manufacturer's instructions. Participants warmed up for five minutes in a cycle ergometer at a comfortable self-selected cadence and then were positioned on the equipment with trunk and thighs firmly strapped and hips flexed at 85° (0°=anatomic position). A specific warm up consisting of 10 isokinetic repetitions 120°/seg was performed. Three trials of 3-s maximal isometric voluntary contractions were performed at knee angle of 60° (0°=knee fully extended), with 180 s of rest between trials. All participants were instructed to avoid any countermovement before knee extension and were encouraged to perform the test “as hard and as fast as possible” (Maffiuletti et al. 2016) The contraction with highest peak torque was used for further analysis. All tests were conducted by the same investigator (R.L.N.R). Quadriceps femoris specific tension was calculated as the ratio between (1) 1RM and QF_{MT} and (2) PT and QF_{MT}.

Body composition

Total body mass, fat-free mass, fat mass and bone mineral content were obtained using imaging with dual-energy X-ray absorptiometry (GE Healthcare Lunar, model Lunar Prodigy Madison, USA) which was calibrated once a day according to the manufacturer's recommendations. Participants were instructed to wear light clothes to remove any metallic material or similar accessories. They were positioned in supine position, lying still during total body scanning (approximately 8 min) and the results were retrieved from the equipment's software (Encore version 14.1, Lunar Prodigy Madison, USA). The same researcher (J.L.T) performed all measurements.

Adjustment of protein intake per meal

After PRE assessments, both groups were instructed to consume at least 20 g of high quality protein from whole food sources at each main meal of the day (i.e. breakfast, lunch and dinner) to maximize muscle protein synthesis (Moore et al. 2009, 2015; Mamerow et al. 2014). Participants received guides with examples about combinations of foods that reached the protein requirement per meal and were instructed to follow this recommendation until POST assessments. Instruction guides were given according to individual food preferences of NV and LOV with or without meat, respectively, considering egg, milk, dairy and vegetal protein food sources. Other than these specific adjustments, participants were instructed to not change their eating habits.

Dietary intake control

Dietary intake (total energy, carbohydrate, lipids and absolute and relative protein intake) and estimated energy availability (total energy minus energy expenditure with physical exercise divided by fat free mass) was obtained from 24-hour Dietary Recall interviews (R24h) (Castell et al. 2015) conducted by the study nutritionists (G.L.M and B.F.F). R24h reflected a weekday and was performed four times during the study. The first interview (1st24h) was conducted before any nutritional instruction was given to the participants to assess their baseline dietary intake. The second (2ndR24h) and third R24h (3rdR24h) interviews were performed during the intervention at weeks 4 and 8, respectively. The last one (4thR24h) was conducted at POST to assess dietary intake and adherence to dietary recommendations by the end of the study.

Adherence to the dietary recommendations was assessed and categorized in agreement to a percentage of adequacy of three main meals, at 2ndR24h, 3rdR24h and 4thR24h. Protein consumption at breakfast, at lunch and at dinner was individually analyzed and meals that reached a quantity equal or greater than 20 g or 0.25 g/kg were considered “adequate”. Each follow-up R24h were categorized as “without”, “low”, “moderate” or “high” adherence when no (0%), one (33.3%), two (66.6%) or all three (100%) main meals of the day were adequate to the dietary recommendation, respectively. Dietary intake of all R24h was calculated using Nutrition Support Program - Nutwin® (Federal University of São Paulo - Escola Paulista de Medicina, Brazil).

Statistical analysis

Continuous data are presented as mean ± standard deviation or [mean (95% confidence intervals)], and categorical data are expressed as frequencies and percent values. The Chi-squared test of homogeneity of proportions was used to assess differences between groups regarding adherence to dietary recommendations. A one-way analysis of variance for repeated measures adjusted for mixed models was used to assess the primary outcomes (skeletal muscle parameters), as well as the secondary outcomes (dietary intake and body composition). For all muscular and anthropometric variables, the main effects of group (LOV vs NV), time (PRE vs POST) and the group x time interactions were assessed. For dietary intake variables, the main effects of group (LOV vs NV), time (1^{st},2^{nd},3^{rd} and 4^{th}R24h) and the group x time interactions were also tested. Mixed model assumptions were tested using residual analysis. Relative percentage change was calculated using the following equation: [(Post values/Pre values - 1) × 100]. Mann-Whitney U test was used to compare the total volume load of leg press and knee extension exercises between groups. Significance was set at $\alpha = 0.05$ and statistical analyses were performed using SPSS software (v. 25, IBM SPSS Inc., USA). Effect sizes (Cohen's $d = (M2 - M1)/SD_{pooled}$, were $SD_{pooled} = \sqrt{[(SD12 + SD22)/2)]}$; and M2 and M1 were means of LOV and NV groups respectively) were calculated and classified as trivial ($\leq$ 0.19), small (0.20 to 0.49) moderate (0.50 to 0.79), large (0.8 to 1.29) and very large ($\geq$ 1.30) effect, according to previous publications. (Espirito Santo and Daniel 2015)

RESULTS

Participants

From the 26 LOV and 38 NV who took part in the intervention, 17 LOV (26.9 ± 6.7 years; 22.5 ± 2.4 kg/m²; 13 female) and 27 NV (28.4 ± 10.11 years; 23.6 ± 2.4 kg/m²; 16 female) completed the study. LOV were vegetarians for an average of 5 years [68.6 (36.6 – 100.6) months]. Both groups completed the training programs with no difference in knee extension and leg press volume-load between groups ($p>0.05$, Figure 2A and 2B, respectively). No adverse event related to this study is reported.

Skeletal Muscle Thickness

Muscle thickness of the quadriceps femoris was similar between groups at baseline and both groups increased muscle thickness of the RF ($p=0.02$), VI ($p<0.01$), VM ($p<0.01$) and VL ($p<0.01$) after training (Table 2)[1]. Accordingly, QF_{MT} increased at POST ($p<0.01$), but no differences were observed between LOV and NV ($p>0.05$) (Figure 3).

Muscle Strength and Specific Tension

Knee extension 1RM and isometric peak torque increased ($p<0.01$) similarly in both groups in the POST assessment period (Figure 4a and 4b, respectively). No main group effect ($p>0.05$) nor main time × group interaction ($p>0.05$) were found (Table 2; also see supplementary file, Table S1 and Table S2).

Quadriceps femoris specific tension calculated from both strength measures (i.e. PT and 1RM) was similar between groups at baseline ($p>0.05$). Both groups showed a similar increase in skeletal muscle specific tension after training ($p<0.01$; Table 2).

Body composition

An increase in participant's total body mass and fat-free mass was observed after training, whereas fat mass decreased ($p<0.05$). No differences were observed between groups ($p>0.05$) and bone mineral density remained unchanged ($p>0.05$) throughout the study. Body composition data are shown in Table 2.

Dietary intake and adherence to nutritional adjustment

Total energy and carbohydrate intake did not differ between groups and remained unchanged throughout the study ($p>0.05$), but lipid and protein consumption was less in the LOV group ($p<0.05$) at baseline (Table 3). Protein intake relative to body mass also differed at baseline (LOV = 0.9 ± 0.5; NV = 1.3 ± 0.5 g/kg) and although both groups increased their protein ingestion after receiving individual recommendations, the difference between LOV and NV was still significant at the end of the intervention

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($p<0.05$) (Table 3). The energy availability did not differ between groups and remained unchanged at 1^{st} and 4^{th}R24h (LOV 41.8 ± 18.4 and 46.8 ± 16.9 kcal/kg FFM; NV 39.2 ± 18.2 and 44.5 ± 17.1 kcal/kg FFM, respectively; $p>0.05$).

Adherence to the dietary recommendations is summarized in Table 4. There was no significant effect of association between groups and the categories of adequacy to dietary recommendation in 2^{nd}R24h, 3^{rd}R24h and 4^{th}R24h ($p=0.13$; $p=0.10$; $p=0.72$, respectively). However, both groups had higher frequency of adequacy at “moderate” and “high” categories in follow-up R24h compared to baseline. No individual was classified as “without adequacy” throughout the study.

DISCUSSION

The present study aimed to compare skeletal muscle and body composition adaptations following 12 weeks of ST in LOV and NV adults with adjusted protein intake per meal. Despite marked differences in protein source and significant lower protein intake by LOV, a similar increase in strength, muscle mass and improved body composition was observed between groups. This suggests that with proper protein intake, exclusion of meat from diet does not impair skeletal muscle and body composition improvements at early stage of ST.

Changes in muscle mass are determined by the net balance between muscle protein synthesis and breakdown (Phillips 2014), which in turn are influenced by skeletal muscle load (Kumar et al. 2009; Mijnarends et al. 2018) and amino acid supply (Moore et al. 2009; Moore 2019). An important question addressed in the current study was whether lack of meat consumption would pose a limitation to skeletal muscle hypertrophic adaptations in LOV. Interestingly, both groups showed similar increases in muscle mass, as reflected by improvements in QF_{MT} and fat-free mass. Our results agree with findings from a recent cross-sectional study by Boutros et al. 2020, reporting no difference in total lean body mass between vegans and omnivores, despite lower protein intake in the vegan group (i.e. 1.11 vs 1.45 g/kg of body mass). Therefore, the current findings expand on previous cross-sectional work by suggesting that a LOV diet is not detrimental for resistance training-induced skeletal muscle hypertrophy in previously untrained individuals.

In the present study, LOV and NV showed similar increments in maximal strength and specific tension. Previous longitudinal works investigated the effect of switching to a meat-free diet on ST adaptation in older adults (Campbell et al. 1999; Wells et al. 2003; Haub et al. 2005). For example, the work by Haub, Wells and Campbell (2005) and Campbell et al. (1999) observed a similar significant increase in muscular strength between individuals who ate meat and those who stopped eating meat after taking part in their studies. Nonetheless, it is important to consider that the aforementioned studies included older non-vegetarian individuals who excluded meat intake during the intervention period. This differs from long-term adherence to a lacto-ovo-vegetarian diet since switching nutritional patterns during the study may have also influenced their results. Although the lack of difference in dynamic and isometric strength may relate to the relatively large neural contribution to strength adaptations at early stages of ST, the current and previous studies agree that either long-term lacto-ovo-vegetarian diet or short-term change to a meat-free dietary pattern does not impair initial maximal strength gains to ST.

The training program also improved participants' body composition, with similar increase in body fat-free mass and decrease in fat mass. The absence of changes in bone mineral content in both groups was an expected result, as improvements are usually observed with longer training periods (~12 months) (Layne and Nelson 1999; Almstedt et al. 2011; Westcott 2012). Accordingly, Westcott et al. (2009) showed that two training sessions per week were effective to improve body composition. In the present study, self-reported energy intake and energy availability did not change throughout the study, thus the increased energy expenditure from adding ST to the participants routine might explain the decreased fat mass (Heden et al. 2011).

To note, LOV and the NV groups only partially adhered to the study intake recommendations. Nevertheless, an increase in daily protein consumption was observed in both groups during the experimental period an at level preconized to support ST adaptations (i.e. 1.2 to 2.0 g/kg/day) by the American College of Sports Medicine (Thomas et al. 2016) and the International Society of Sports Nutrition (Kerksick et al. 2018). This increase may be explained by adjusted protein intake per meal, which promoted the consumption of ~ 60 g of protein per day beyond to the amount of protein consumed from other daily meals.

Even though NV consumed more protein (ranging from 1.6 up to 1.8 g/kg/day) compared to LOV (1.3 up to 1.4 g/kg/day) skeletal muscle adaptations were similar between groups, which may be counterintuitive since greater protein ingestion is thought to further stimulate skeletal muscle protein synthesis and potentiate training adaptations (Schoenfeld et al. 2013; Jäger et al. 2017; Morton et al. 2018; Nunes at al. 2022). A recent study conducted from Hevia-Larrian et al. (2022) showed that people on non vegetarian and vegan diets had similar increments in muscle mass after 12 weeks of ST, but the intervention protocol used dietetic supplementation to ensure both groups reached the same protein intake (i.e. 1.6 g/kg of body mass). In view of the similar protein intake, it was more plausible that differences in hypertrophic increments were not observed in their study, mainly because a high plant based protein intake can stimulate a great muscle protein synthetic responses (Pinckaers et al. 2021).

The difference in total daily protein intake between LOV and NV was not surprising, because meat is a great dietary source of protein and, in the present study, supplementation was not used to compensate for protein intake. For example, one medium size beef stake (ie. 100g) contains ~27g of protein, while 100g of egg (2 units) or milk (1/2 cup) provides, respectively, 12.4 and 3.3 g of protein (USDA, 2021). Although there are few studies assessing anabolic properties of food sources, meat, milk and eggs are considered high quality protein sources owing to their large amount of essential amino acids, which can increase myofibrillar protein synthesis rates in young men during recovery from exercise (Burd et al. 2015; Van Vliet et al. 2017; van Vliet et al. 2018). It was also considered that vegetarian diets are based in plant food sources and, although LOV group also have consumed egg and dairy products, vegetable protein is known to have a lower quality (van Vliet et al. 2015, 2018; Berrazaga et al. 2019). Nevertheless, the vegetable amino acids profile can be balanced with adjustments such the combination of cereals and legumes in a same meal, which was also encouraged by the researchers (Berrazaga et al., 2019; Van Vliet et al., 2018). In addition, vegetable have a rich food matrix and the ingestion of whole plant food sources includes not only protein, but different amounts of other macronutrients, vitamins, minerals and phytochemicals, which besides of stimulating muscle protein remodeling may also provide additional muscle recovery and health benefits (D'Angelo, 2020; Fuhrman and Ferreri, 2010; Van Vliet et al., 2018).

One possible limitation of the present study was the use of 24-hour recall record to assess participants' food intake. It could be argued that a three-day food record would be more effective to represent individual food habits, but the 24-hour recall is a valid method and it does not depend on the registration made by the participant, who may forget and/or have difficulties to complete it themselves. Additionally, it was not possible to assess and compare protein quality between groups, which is a limitation of the study given that high-quality protein is thought to bring about greater MPS. Even with limitations, these findings contribute to the body of knowledge in this area, which has been overlooked despite the growing number of vegetarians worldwide. In addition, the present study prioritized the protein intake through food sources without use of supplements, which has great applicability to daily life. The current findings may suggest that during early training stages of ST, protein intake near 1.2-1.4 g/kg/day may suffice to support anabolic adaptations to ST in untrained individuals taking in a lacto-ovo-vegetarian diet. Nevertheless, it is possible that longer training periods (i.e. beyond 3 months) may be needed any difference in daily protein intake to be translated into distinguishable hypertrophic gains between groups. Further studies (1) employing longer interventions (2) with trained LOV and NV individuals and (3) assessing differences in protein quality between groups will be needed to test this hypothesis.

CONCLUSION

Lacto-ovo-vegetarians and non-vegetarians showed similar improvement in maximal strength, muscle mass, and body composition to 12 weeks of ST, despite large differences regarding protein source and intake. Although there is still a gap regarding how different food sources impact on ST adaptations, our results showed that following a lacto-ovo-vegetarian diet does not pose a limitation for early-stage ST adaptations in previously untrained individuals. Furthermore, meat consumption and protein intake higher than the minimum recommended for individuals engaged in exercise (1.2 g/kg/day), did not provided additional benefits compared to a lacto-ovo-vegetarian diet.

CONFLICTS OF INTEREST

The authors declare there are no conflicts of interest

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AUTHORS' CONTRIBUTIONS TO THE MANUSCRIPT

GLM, RSP and CGS designed research;

RSP and CGS provided essential materials;

GLM, CMB, BFF, MLO, RLN, FLC and JLT analyzed data or performed statistical analysis;

GLM, RSP, CMB, ENW, JLT and CGS wrote the paper;

GLM, RSP, CMB, BFF, MLO, RLN, FLC, ENW, JLT, CGS had primary responsibility for final content;

All authors (GLM, RSP, CMB, BFF, MLO, RLN, FLC, ENW, JLT, CGS) read and approved the final manuscript.

DATA AVAILABILITY STATEMENT

Data generated and analyzed during this study are available from the corresponding author upon reasonable request.

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Table 1. Strength training program

Weeks	Sets	Repetitions	Intensity	Rest between sets
1-3	2	15-12	15-12RM	30 s – 1 min
4-6	2	12-10	12-10RM	1 min and 30 s
7-9	3	8-10	8-10RM	2 – 3 min
10-12	3	6-8	6-8RM	3 – 4 min

RM= repetition maximum

Table 2. Skeletal muscle parameters and body composition of LOV and NV during the study

	LOV		**NV**	
	PRE	**POST**	**PRE**	**POST**
Muscle Thickness				
RF (mm)	20.1 ± 3.2	21.1 ± 4.1†	21.4 ± 3.2	22.9 ± 4.2†
VI (mm)	16.5 ± 3.6	19.3 ± 3.3†	15.9 ± 3.6	18.0 ± 3.4†
VM (mm)	20.6 ± 4.2	23.8 ± 4.1†	21.6 ± 4.2	23.8 4.3†
VL (mm)	21.8 ± 3.2	23.4 ± 2.9†	23.1 ± 3.2	23.9 ± 3.0†
Maximal strength tests				
1RM (kg)	84.2 ± 18.7	117.9 ± 12.4†	87.1 ± 18.7	114.9 ± 12.1†
PT (N·m)	207.0 ± 44.0	268.4 ± 27.1†	239.5 ± 44.0	263.9 ± 26.6†
Body composition				
Total body mass (kg)	62.3 ± 10.6	63.1 ± 8.9†	66.8 ± 10.6	67.4 ± 9.4†
Fat-free mass (kg)	38.3 ± 8.6	39.5 ± 7.3†	42.3 ± 8.6	43.7 ± 7.7†
Fat mass (kg)	21.7 ± 5.9	21.1 ± 4.7†	22.0 ± 5.9	21.3 ± 4.9†
BMC (kg)	2.3 ± 0.4	2.3 ± 0.3	2.4 ± 0.4	2.4 ± 0.3

Mean ± standard deviation; † main time effect ($p < 0.05$); BMC= bone mineral content; LOV= lacto-ovo-vegetarian; NV= non-vegetarian; POST= 13week; PRE= 0week; PT= isometric peak torque; QF= overall muscle thickness of the quadriceps femoris; RF= rectus femoris; VI= vastus intermedius; VL= vastus lateralis; VM= vastus medialis; 1RM= one-repetition maximum knee extension test.

Table 3. Adjusted mean of dietary intake of LOV and NV at each twenty-four hour dietary recall interview

	LOV				NV			
	1stR24h	**2ndR24h**	**3rdR24h**	**4thR24h**	**1stR24h**	**2ndR24h**	**3rdR24h**	**4thR24h**
Dietary intake								
Energy (Kcal)	1808.2±789.0	1686.7 ± 494.4	1774.3 ± 750.4	2060.6 ± 724.8	1877.8 ± 798.1	2091.1 ± 498.2	2322.1 ± 776.8	2199.2 ± 730.6
Protein (g)	55.2 ± 35.0	76.3 ± 28.3†	73.7 ± 29.6 †	87.5 ± 42.8†	85.1 ± 35.0*	104.8 ± 28.9† *	114.0 ± 30.5 † *	121.9 ± 42.9 † *
Protein (g/kg)	0.9 ± 0.5	1.2 ± 0.4†	1.2 ± 0.5 †	1.4 ± 0.6†	1.3 ± 0.5 *	1.5 ± 0.4 † *	1.7 ± 0.5 † *	1.8 ± 0.6 † *
Carbohydrate (g)	271.3 ± 112.6	211.0 ± 79.6	239.8 ± 85.2	286.7 ± 123.6	231.5 ± 112.6	235.6 ± 80.0	251.3 ± 87.7	248.5 ± 124.5
Lipids (g)	60.2 ± 33.3	62.3 ± 29.1	58.1 ± 43.5	66.5 ± 21.2	68.0 ± 33.3*	74.7 ± 22.0*	90.9 ± 45.2 *	78.2 ± 21.5*

EA: energy availability; FFM: fat-free mass (kg); Mean ± standard deviation; † = main time effect ($p < 0.05$); *main group effect ($p < 0.05$); LOV= lacto-ovo-vegetarian; NV= non-vegetarian; 1stR24h= first 24hour dietary recall interview applied; 2ndR24h= second 24hour dietary recall interview applied; 3rdR24h= third 24hour dietary recall interview applied; 4thR24h= forth 24hour dietary recall interview applied.

Table 4. Frequency diet adherence adequacy of the three main meals in the day throughout the study in LOV and NV groups

Category of adequacy	**Percent of adequacy of three main meal in the day**					
	2nd R24h		**3rd R24h**		**4th R24h**	
	LOV	**NV**	**LOV**	**NV**	**LOV**	**NV**
Without	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Low	2 (12.5%)	2 (7.7%)	3 (17.6%)	1 (3.7%)	0 (0%)	1 (3.7%)
Moderate	10 (62.5%)	9 (34.6%)	8 (47.1%)	9 (33.33%)	9 (52.9%)	11 (40.7%)
High	4 (25.0%)	15 (57.7%)	6 (35.3%)	17 (63.3%)	8 (47.1%)	15 (55.6%)

Absolute frequency (relative frequency); †= significant positive association ($p < 0.05$); High=100%; LOV= lacto-ovo-vegetarian; Low= 33.3%; Moderate= 66.6%; No= 0%; NV= non-vegetarian; 2nd R24h = second 24hour dietary recall interview applied; 3rd R24h = third 24hour dietary recall interview applied; 4th R24h = forth 24hour dietary recall interview applied.

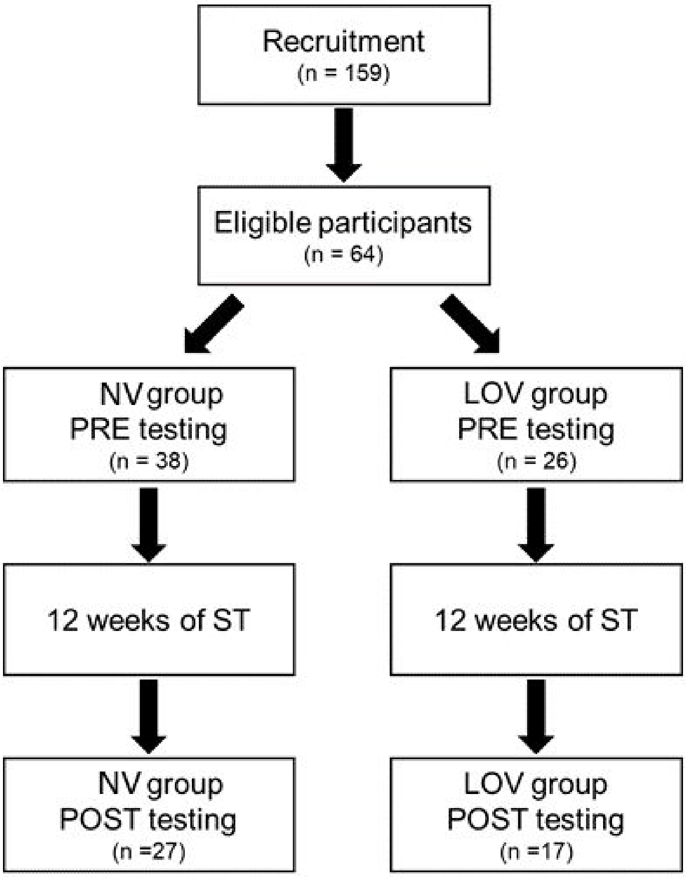

Figure 1. Flowchart showing the experimental design of the study. Participants in the non-vegetarian (NV) and lacto-ovo-vegetarian (LOV) groups were tested a baseline (PRE) and testing and after (POST) 12 weeks of strength training (ST) intervention.

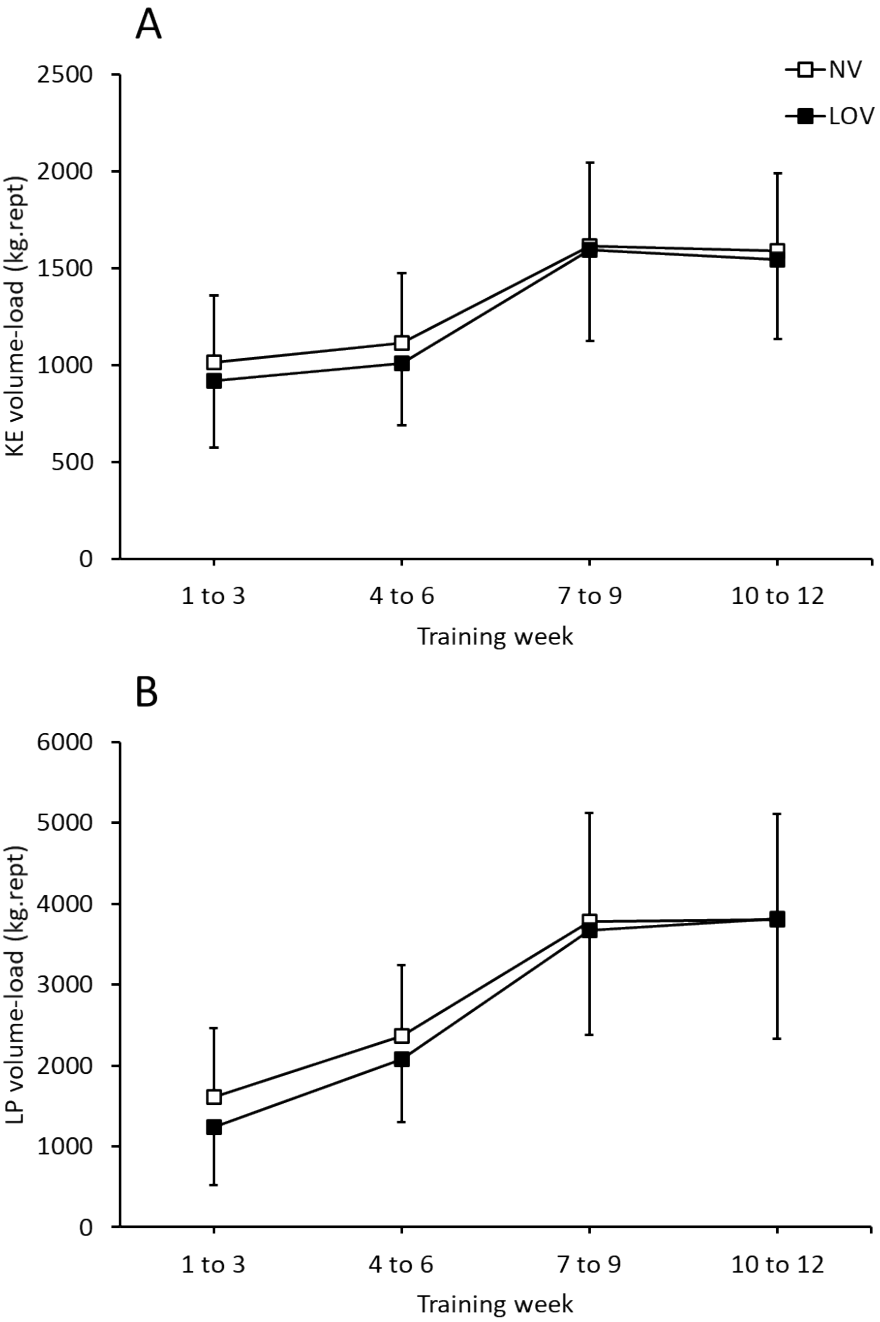

Figure 2. Mean knee extension (KE, panel A) and leg press (LP, panel B) volume-load per mesocycle in the non-vegetarian (NV) and lacto-ovo-vegetarian (LOV) groups. No differences in volume-load were observed between groups and both NV and LOV increased volume-load as training progressed (data not shown).

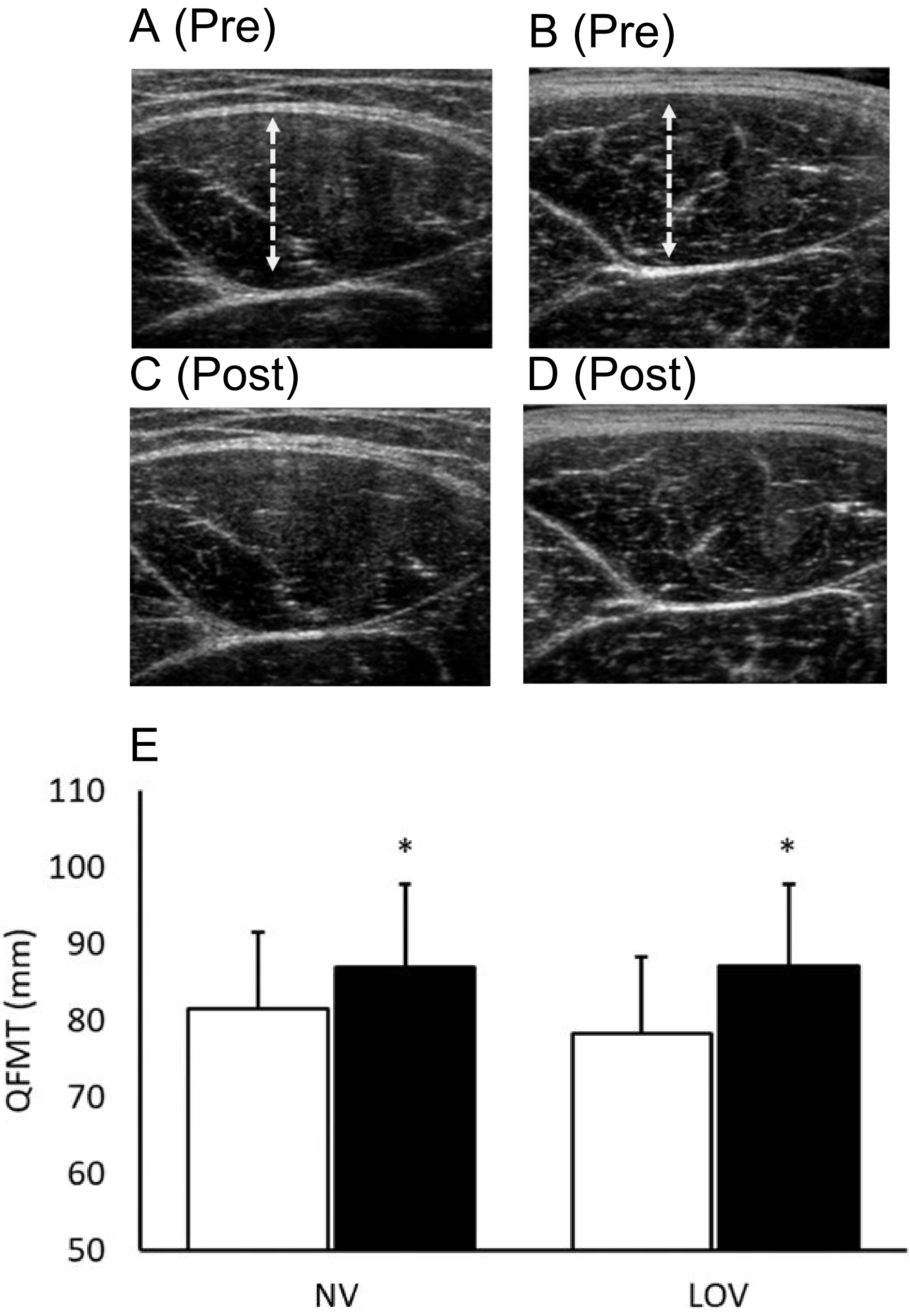

Figure 3. Illustration of quadriceps femoris muscle thickness (top panels) and quadriceps femoris muscle thickness (QFMT – bottom panel) of non-vegetarian (NV) and lacto-ovo-vegetarian (LOV) before and after training. Panel A and C depicts recording of a typical NV before and after training, respectively. Panel B and D depicts LOV images before and after training, respectively. An increase in QFMT post training was observed in both groups ($p<0.05$), with no differences between groups (main group effect and time*group interaction $p>0.05$). * = significance time-effect ($p<0.05$).

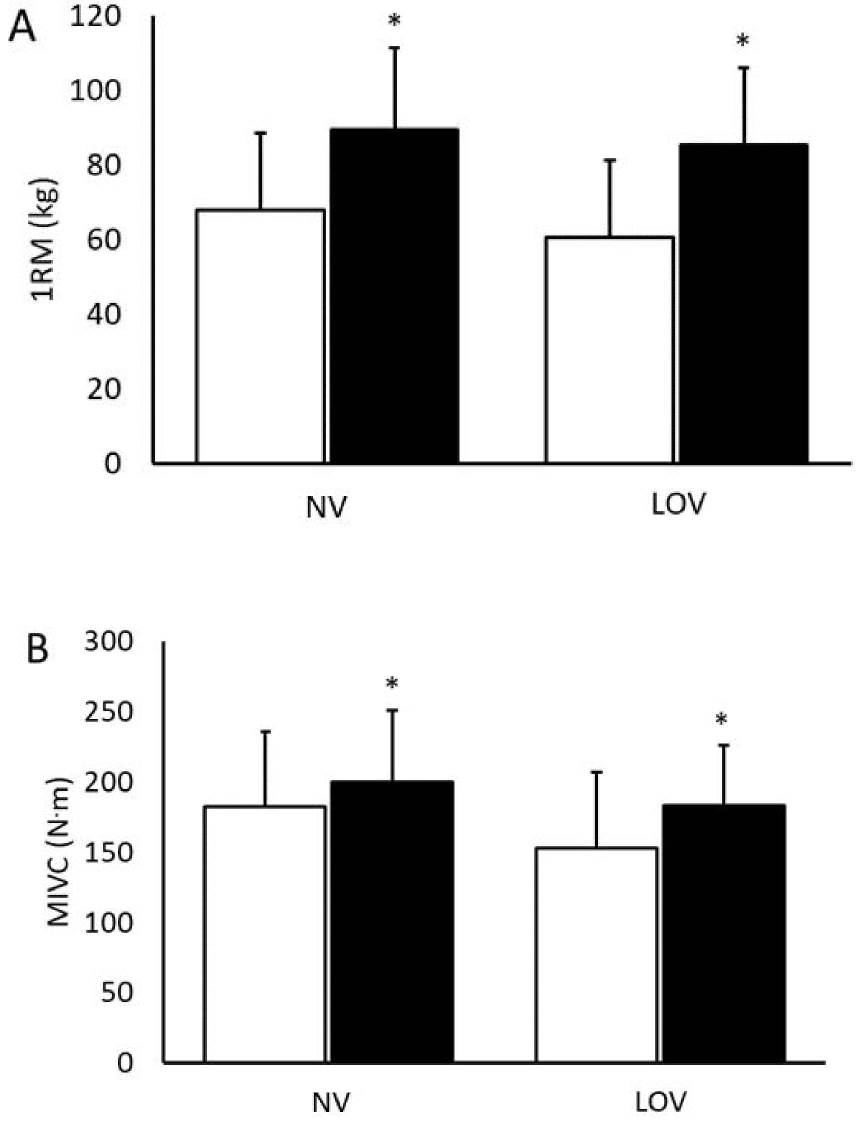

Figure 4. Knee extension 1-repetition maximum (1RM) (panel A) and maximal isometric voluntary contraction (MIVC) peak torque (panel B) in non-vegetarian (NV) and lacto-ovo-vegetarian (LOV) before and after training. Maximal strength increased in both groups ($p<0.05$) post training, with no difference between groups (main group effect and time*group interaction $p>0.05$). * = significance time-effect ($p<0.05$).