

The Impact of Long-Chain Omega-3 Polyunsaturated Fatty Acid Supplementation on Body Composition, Strength, and Power in Collegiate Athletes

Original Research

Jeffery L. Heilesen¹, Ashlyne Elliott², Julie A. Buzzard¹, Mitchell C. Cholewinski¹, Andrew Gallucci¹, LesLee K. Funderburk¹

¹Baylor University, Waco, TX/USA

²Veterans Health Administration, Houston, TX/USA

Open Access

Published:
February 28, 2022



Copyright, 2022 by the authors. Published by Pinnacle Science and the work is licensed under the Creative Commons Attribution 4.0 International License. To view a copy of this license, visit <http://creativecommons.org/licenses/by/4.0/>

Journal of Exercise and Nutrition: 2022, Volume 5 (Issue 1): 5

ISSN: 2640-2572

Abstract

Introduction: Recent evidence suggests that long-chain omega-3 polyunsaturated fatty acid (LC n-3) supplementation may enhance training adaptations associated with athletic performance. This study examined the impact of LC n-3 supplementation on body composition, strength, and power in collegiate athletes.

Methods: Athletes (n = 27) were assigned to one of two conditions for eight weeks: fish oil (FO, 3.0 g·d⁻¹ [1.75g EPA and 1.1g DHA], n = 15) or placebo (PL, high-oleic safflower oil, 3g, n = 12) for 8-weeks. Athletes completed a three-day food log and questionnaire, provided a blood sample via fingerstick to determine their LC n-3 status, conducted body composition analysis through dual energy x-ray absorptiometry, and had their handgrip strength (HGS) and countermovement jump assessed.

Results: In the FO group, the omega-3 index, EPA and DHA increased by 73%, 332% and 64%, respectively, while there was no change in the placebo group. HGS significantly improved in the FO group ($p = .018$, +9.1%) and did not change in the placebo group ($p = .615$, -1.8%). Body composition and power were similar between groups. The change in HGS was positively correlated with the relative change in EPA and EPA:AA ratio.

Conclusions: For in-season athletes, the addition of LC n-3 supplementation to a dietary regime increases blood LC n-3 status and may preserve or improve muscular performance while in-season.

Key Words: athletic performance, eicosapentaenoic acid, docosahexaenoic acid

Corresponding author: LesLee K. Funderburk, leslee_funderburk@baylor.edu

Introduction

Long-chain omega-3 polyunsaturated fatty acids (LC n-3), primarily the bioactive constituents eicosapentaenoic acid (EPA, 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3), are widely recognized for their cardio- and neuroprotective capabilities^{1,2} and have recently been associated with some aspects of athletic performance and recovery.³⁻⁵ The influence of LC n-3s on various physiological

processes appears to be mediated by its incorporation into tissue phospholipid membranes.⁶ Mechanistically, increasing skeletal muscle LC n-3 membrane content may improve insulin sensitivity and enhance anabolic signaling pathways.^{7,8} A seminal study by Smith et al.⁹ has shown that LC n-3 supplementation of 4g·d⁻¹ (3.36g EPA+DHA) for 8-weeks alters skeletal muscle fatty acid composition and increases muscle protein synthesis in young and middle-aged adults. Additionally, EPA and DHA have been shown to reduce markers of head trauma^{10,11} and improve cardiovascular risk factors in athletes.¹² As such, LC n-3 supplementation, typically as fish oil (FO), has been suggested as a safe and effective means to improve athlete health and performance.

Researchers suggest that athletes do not consume adequate preformed EPA and DHA from dietary sources, such as fatty fish. Similarly, athletes also have sub-optimal levels of erythrocyte EPA plus DHA (as a percentage of total fatty acids), also referred to as the Omega-3 Index (O3i).^{13–16} Within the context of training, recent biomarker studies have concluded that training may deplete LC n-3 status, as measured by the O3i, and leads to greater accumulation of n-6 fatty acids, namely arachidonic acid (AA; 20:4n-6), within the tissue in the absence of consistent fish intake or supplementation.^{17–19} Tissue incorporation of LC n-3 occurs at the expense of n-6 fatty acids and has been associated with a reduction in inflammatory cytokines and reactive oxygen species in athletes.²⁰ The EPA:AA ratio has recently been reported as an indication of this process and has also been implicated as a potential predictor of cardiovascular disease risk.²¹ Additionally, LC n-3 depletion may also have negative implications for physical performance in athletes, especially since whole blood and plasma EPA and the EPA:AA ratio are correlated with strength.^{16,22} Our recent cross-sectional analysis found that handgrip strength was associated with whole blood EPA and the EPA:AA ratio and negatively correlated with the n-6:n-3 ratio in athletes.¹⁶ Hence, LC n-3 supplementation, through the incorporation of EPA and DHA into the skeletal muscle phospholipid and at the expense of n-6 fatty acids, may enhance strength, power and promote lean body mass (LBM) accretion in athletes.

Accordingly, the primary purpose of this investigation was to determine the effect of LC n-3 supplementation, via FO, on the promotion of lean body mass, strength, and power in athletes during in-season training. The secondary aim of this study was to determine if the change in strength and power are related to the change in whole blood LC n-3 fatty acid status. We hypothesized that LC n-3 supplementation would increase LBM, strength, and power during in-season and that the changes in strength and power will be associated with the change in whole blood fatty acid status.

Scientific Methods

This study was a randomized, double-blind, placebo-controlled trial in which participants were randomly assigned by a computer-generated sequence to either receive 3g·d⁻¹ FO (1.75g EPA, 1.10 g DHA) or 3g·d⁻¹ safflower oil (PL), both consumable in 5 soft gel capsules. Participants were instructed to maintain consistent dietary and physical training habits throughout the study. Three-day food records, including an n-3 food frequency questionnaire (FFQ), were collected at the beginning and end of the study. Participants had a blood sample collected via a finger stick, underwent body composition analysis, and completed strength and power assessments. Assessments were collected on week one and at the end of the 8-week study at approximately the same time of day.

Participants

Thirty-six (20 ± 1.5 y) collegiate athletes volunteered for the study (n = 14 males, n = 22 females). Twenty-seven (77%) completed the intervention. Athletes were recruited from the NCAA D1 teams, intramural clubs, and the university competitive dance team. All athletes volunteered for the study during their sports' respective in-season. Participants were screened and excluded if they had any medical conditions that limited training. The study was approved by the University's Institutional Review Board and written informed consent was obtained from each participant.

Three-day Food Record and Omega-3 Food Frequency Questionnaire

To assess macronutrient intake before and after the intervention, athletes tracked all calorie containing foods and beverages three days (two weekdays and one weekend) via a smartphone app (MyFitnessPal®). This mobile application has been validated as an effective method to measure self-reported dietary intake.²³ Additionally, to specifically determine their habitual intake of omega-3 fatty acids, athletes were asked to complete a validated 21-item omega-3 FFQ.²⁴ The FFQ quantifies the omega-3 intake from plant and marine sources based on the frequency and portion size consumed over the last day, week, and 6 months.

Supplementation Protocol

Participants in the FO (1.75g EPA, 1.10g DHA) and PL (3g high-oleic safflower oil) groups were instructed to consume 5 capsules per day of their assigned supplement. The athletes were asked to take their supplement with a meal

containing fat, typically the evening meal, to maximize absorption. Participants were provided their supplements in two week increments. The FO supplementation contained a higher EPA dose than DHA, which was selected due to the pre-clinical data identifying EPA as the primary n-3 component involved in muscle protein turnover.²⁵ Both the treatment and placebo products were produced from CAS BioSciences, LLC (New York USA / California USA). The FO and PL products used in this study were manufactured under the applicable US Food & Drug Administration Good Manufacturing Practices, and were packaged for this study by CAS BioSciences, LLC's partner company, Gemini Pharmaceuticals, Inc. (New York USA), under US Food & Drug Administration 21 CFR Part 211 pharmaceutical standards.

Body Composition

Body mass was measured on a mechanical scale while the participant wore light, athletic clothing without shoes and their height was measured with a stadiometer (Seca 703, China). Body composition (LBM and fat mass [FM]) was estimated using the Dual Energy X-ray Absorptiometry (DXA, Discovery DXA™, Hologic®, Bedford, MA). The fat-free mass index (FFMI), a height adjusted measure of lean body and bone mass, was calculated by dividing the FFM by height squared²⁶ and used for analysis.

Strength and Power

Handgrip strength (HGS) was measured using a dynamometer (Hydraulic Hand Dynamometer, Baseline® Evaluation Instruments) at baseline and at the end of the study. The participants were instructed to hold the dynamometer with their dominant arm with their elbow at a 90° angle and squeeze as hard as possible until the observer started to see a decrease in the reading. The width of the dynamometer was adjusted for each participant to ensure a firm grip. This was indicated by the phalanx positioned 90° while covering the handle. The attempt was recorded in kilograms and the gauge was reset. The athlete rested at least one minute between attempts. Participants were assessed on three separate attempts and the results were averaged for data analysis.

Vertical jump height was assessed by measuring countermovement jump (CMJ) using the VALD ForceDecks dual-force plates and the information was analyzed with the VALD ForceDecks software (Version 2.0.7782) at baseline and at the end of the study. Peak (PP) and mean power (MP) were calculated from body mass (kg) and jump height (cm) using validated formulas.^{27,28} All athletes conducted CMJ on a regular basis for internal testing, hence, the familiarization session was just prior to the first measure to standardize the process. The participants were instructed to conduct the CMJ with hands on hips to eliminate upper limb technique. The jumps were performed three times allowing for a standard one-minute rest interval in-between each jump. The average of the three attempts were used for data analysis.

Blood Fatty Acid Analysis

A drop of blood was collected from each participant via finger stick on filter paper pre-treated with a cocktail solution (Fatty Acid Preservative Solution, FAPS™) and allowed to dry at room temperature for 15 min. The dried blood spots (DBS) were shipped to OmegaQuant for fatty acid analysis. Fatty acids were identified by comparison with a standard mixture of fatty acids characteristic of RBC (GLC OQ-A, NuCheck Prep, Elysian, MN) which was also used to construct individual fatty acid calibration curves. Fatty acid composition was expressed as a percent of total identified fatty acids. The O3i is defined as the sum of EPA and DHA adjusted by a regression equation ($r = 0.96$) to predict the O3i in the RBC.

Statistical Analysis

Data reported as means and standard deviations. Relative and absolute change scores are reported as mean and 95% confidence intervals (lower bound, upper bound). Baseline and dependent variables with change scores, and percent changes were assessed for group differences via a *t*-test. After determining data normality and homogeneity of variance using the Shapiro-Wilks and Levene's Test for Equality of Variances, a mixed model repeated measures ANOVA was used to determine if interactions between group and time occurred for body composition, strength, or power. For significant findings, multiple comparison testing was performed using a Bonferroni adjustment. Between group differences were further analyzed using effect sizes (Cohen's *d*). The effect sizes (ES) were classified as follows: < 0.20, trivial; 0.20–0.49, small; 0.50–0.79, moderate; ≥ 0.80 , large.²⁹ Pearson correlations were used to examine the relationship between whole blood fatty acids and changes in strength and power. Data were analyzed using SPSS version 27 (IBM SPSS, Chicago, IL). Significance was set *a priori* at $p < .05$. Based on the various performance metrics in a similar population, we estimated that our sample size should be between 24 and 32 athletes ($f = 0.26$ – 0.30 , $1 - \beta = 0.80$, $\alpha = 0.05$).³⁰

Results

Participants

Of the 36 athletes randomized to a group, 27 (77%) completed the intervention. Of the 9 dropouts, 4 from the FO group and 5 from the PL group, 8 were due to >2 missed supplement distributions and one dropout in the FO group was due to nausea attributed to the intervention. The athletes that completed the study were from track and field ($n = 6$), baseball/softball ($n = 5$), spirit team ($n = 11$), volleyball ($n = 2$), crew ($n = 2$), and acrobatics and tumbling ($n = 1$). All athletes underwent body composition analysis ($n = 27$); however, only partial data were available for whole blood fatty acid analysis ($n = 20$), HGS ($n = 14$), CMJ ($n = 25$), FFQs ($n = 19$) and dietary logs ($n = 12$). There were no group differences for participant age ($p = .972$), body mass ($p = .519$), or height ($p = .335$).

Dietary Intake

There were no group differences at baseline for any macronutrient ($p > .05$). From PRE to POST, dietary intake of calories ($p = .952$), protein ($p = .424$), carbohydrates ($p = .894$) and fat ($p = .629$) were unchanged. There were no group by time differences in dietary intake of calories ($p = .467$), fat ($p = .436$), protein ($p = .997$), or carbohydrate ($p = .655$). From PRE to POST, dietary intake of EPA ($p = .770$), DHA ($p = .286$), and ALA ($p = .903$) were similar between groups.

Omega-3 Index and Whole Blood Omega-3 Fatty Acids

Pre-supplementation O3i and whole blood fatty acids were similar between groups. Figure 1 shows the individualized changes of the O3i from PRE to POST in each group. In the PL group, the O3i (4.67% to 4.52%; $p = .731$), whole blood EPA (0.37% to 0.33%; $p = .852$), DHA (2.62% to 2.49%; $p = .549$), and AA (10.50% to 10.75%; $p = .335$) were similar from PRE to POST. In the FO group, the O3i (4.58% to 7.94%; $p < .001$), whole blood EPA (0.41% to 1.77%; $p < .001$) and DHA (2.46% to 4.04%; $p < .001$) significantly increased, while AA (10.74% to 10.00%; $p = .029$) decreased.

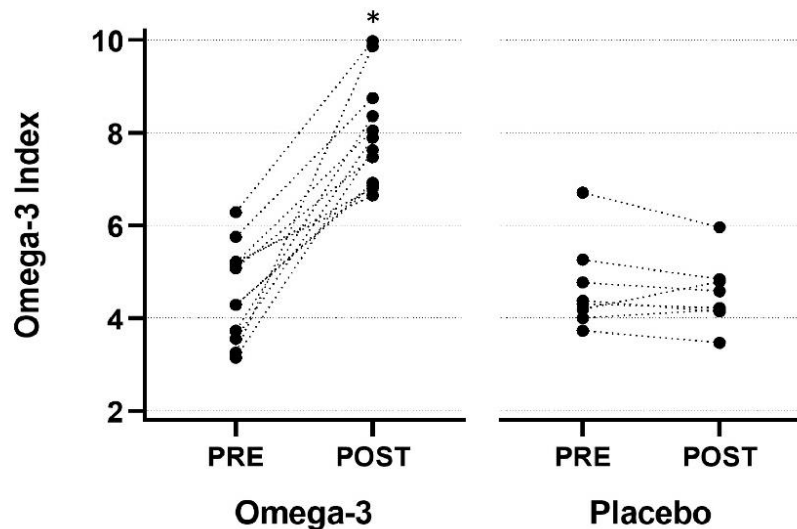


Figure 1. Individual changes in the Omega-3 Index in the fish oil ($n = 12$) and placebo ($n = 8$) groups. Values <4 are high-risk and values >8 are low-risk for cardiovascular disease.

*indicates a significant difference compared to baseline and between groups ($p < .001$).

Body Composition

At baseline, there were no significant group differences in weight, LBM, FFMI, FM, or %BF. Table 1 shows the PRE, POST, and change values of weight, LBM, FFMI, FM, and %BF for each group. Briefly, body composition changes were similar between groups ($p > .05$). The between group effect sizes indicated small differences between LBM ($ES = 0.21$), FM ($ES = 0.38$), and %BF ($ES = 0.47$).

Table 1. PRE, POST, and absolute changes in body composition following 8-weeks of supplementation

	Fish Oil (n = 15)			Placebo (n = 12)		
	PRE	POST	Δ (CI)	PRE	POST	Δ (CI)
Weight (kg)	73.1 ± 14.5	73.5 ± 15.0	0.4 (-0.6, 1.4)	69.7 ± 12.7	70.1 ± 13.8	0.4 (-0.7, 1.5)
LBM (kg)	56.6 ± 14.4	57.3 ± 14.5	0.7 (-0.3, 1.7)	51.9 ± 10.9	52.2 ± 11.9	0.3 (-0.8, 1.5)
FFMI (kg·m ⁻²)	18.9 ± 2.7	19.1 ± 2.9	0.2 (-0.1, 0.6)	18.3 ± 3.2	18.4 ± 3.4	0.1 (-0.3, 0.4)
FM (kg)	13.7 ± 4.6	13.6 ± 4.4	-0.1 (-0.8, 0.6)	15.5 ± 5.6	15.9 ± 6.0	0.4 (-0.4, 1.2)
BF (%)	19.6 ± 7.1	19.3 ± 6.6	-0.4 (-1.3, 0.6)	22.4 ± 6.5	22.7 ± 6.9	0.4 (-0.7, 1.4)

Abbreviations: LBM, lean body mass; FFMI, fat-free mass index; FM, fat mass; BF, body fat.

Data are means ± SD rounded to the nearest 0.1. For absolute changes, 95% confidence intervals (lower bound, upper bound) are reported.

Strength and Power

There was a significant group by time interaction for HGS ($p = .04$). In the placebo group, HGS did not change (PRE: 36.9 ± 10.8 kg, POST: 36.3 ± 11.7 kg; Δ : -0.58 kg [95% CI: $-3.0, 1.9$]; $p = .615$); however, HGS significantly improved from PRE (39.0 ± 14.3 kg) to POST (42.0 ± 14.1 kg) in the FO group (Δ : 3.1 kg (95%CI: $0.6, 5.5$); $p = .018$). The magnitude of the difference indicates a large effect ($ES = 1.33$).

There was no difference in CMJ height from PRE to POST ($p = .806$), between groups ($p = .132$), or between groups over time ($p = .215$). Of note, CMJ height improved in the FO group (PRE: 42.8 ± 7.5 cm, POST: 43.5 ± 7.3 cm; Δ : 0.8 cm [95%CI: $-0.6, 2.1$]; $p = .263$) and declined in the PL group (PRE: 39.2 ± 6.3 cm, POST: 38.7 ± 5.7 cm; Δ : -0.5 cm [95%CI: $-2.1, 1.0$], $p = .499$). The calculated effect size for the difference between groups indicates a moderate effect ($ES = 0.54$). From PRE to POST, PP significantly increased in the FO (3874.1 W ± 1037.6 to 4411.93 W ± 1171.1 ; Δ : **537.8 W** (95%CI: $410.6, 665.0$); $p < .001$) and PL (3446.1 W ± 848.7 to 3924.7 W ± 1025.2 ; Δ : **478.6 W** (95%CI: **335.1, 622.1**); $p < .001$) groups. There was no main effect for group ($p = .282$) or group by time interaction ($p = .529$) noted. The between group effect sizes were small ($ES = 0.25$). No time ($p = .179$), group ($p = .281$), or group by time interaction ($p = .377$) were identified for MP. Figure 2 depicts the percent change in HGS (FO: 9.1% , PL: -1.8%) and CMJ (FO: 2.2% ; PL: -1.1%) between groups.

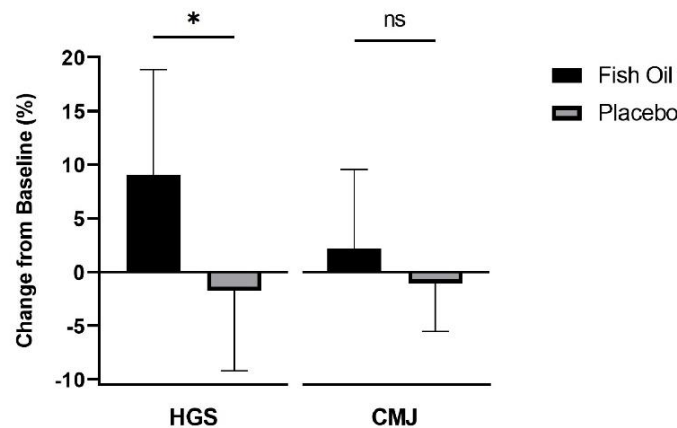


Figure 2. The change from baseline (%) in handgrip strength (HGS) and countermovement jump (CMJ) height in the fish oil (HGS, $n = 7$; CMJ, $n = 14$) and placebo (HGS, $n = 7$; CMJ, $n = 11$) group.

Values are mean and SD. * $p = .038$; ns, $p = .213$

Relationship between Whole Blood Fatty Acids and Strength and Power

The percent change of whole blood EPA ($r = 0.874$, $p = .005$) and EPA:AA ($r = 0.849$, $p = .008$) was positively and strongly correlated with HGS. However, EPA and EPA:AA was not associated with the change in CMJ height ($r = 0.144$, $p = .569$; $r = 0.084$, $p = .740$, respectively), PP ($r = -0.135$, $p = .594$; $r = -0.182$, $p = .469$, respectively), or MP ($r = 0.105$, $p = .679$; $r = 0.078$, $p = .758$, respectively). The percent change of DHA or DHA:AA was not correlated with any performance outcome ($p > .05$).

Discussion

The primary aim of this study was to investigate the effect of fish oil supplementation ($2.85\text{g}\cdot\text{d}^{-1}$ EPA+DHA) for 8-weeks on body composition, strength, and power in collegiate athletes. We demonstrated that LC n-3 supplementation increases HGS and that the relative changes in HGS were positively correlated with the change in whole blood EPA and the EPA:AA ratio. Our results also indicate that LC n-3 supplementation tends to confer favorable effects on body composition and power; however, this relationship is less clear.

We found that LC n-3 supplementation improved HGS in athletes by 3.1 kg (9.1%) over 8-weeks. With or without resistance training, previous trials have reported increases in HGS with LC n-3 supplementation.^{30–32} Using a similar daily dose of EPA+DHA, Lee et al.³⁰ reported that HGS increased by 2.5 kg (9.4%) in combination with resistance training, which was 1.0 kg (4.1%) and 3.5 kg (13.3%) higher than in the resistance training only and control group, respectively. Compared to PL, Smith et al.³¹ reported that FO supplementation improved HGS by 2.3 kg (~6.5%). Conversely, trials providing less potent FO doses do not report beneficial effects on HGS.^{33,34} While we provided $2.85\text{g}\cdot\text{d}^{-1}$ LC n-3 and reported a significant improvement in HGS, our study was the first to demonstrate these findings in young athletes. Notably, Gravina et al.³⁵ found that LC n-3 supplementation ($0.1\text{g}\cdot\text{kg}^{-1}$, $6.3 \pm 1.8\text{g}\cdot\text{d}^{-1}$), in elite soccer players, did not increase 1RM leg extension compared to PL. Although speculative, one plausible explanation for the divergent findings may be that we opted for a field expedient isometric measure compared to a more common 1RM assessment. Previous studies have reported that LC n-3 supplementation may not improve 1RM, but improves isometric strength, which suggests that the effects of LC n-3 are related to muscular, not neural, adaptations.^{36,37} It should be noted that a recent trial, in contrast to Gravina et al., demonstrated that healthy young adults taking $1.4\text{g}\cdot\text{d}^{-1}$ LC n-3 for 4 weeks significantly improved 1RM leg extension (+14.1 kg), whereas there was no improvement in the PL group.³⁸ Interestingly, we noted that the relative change in EPA and EPA:AA was significantly correlated to the change in HGS, while DHA and DHA:AA were not correlated. Our results align with recent studies showing that upper body strength was associated with plasma and whole blood EPA and EPA:AA.^{16,22}

In the FO group, CMJ height increased by 2.2%, while CMJ decreased by 1.1% in the PL group. Notably, the difference was not significant from PRE to POST or between groups, despite the magnitude being indicative of a moderate effect ($ES = 0.54$). From a practical standpoint, the minimal improvement (< 1 cm) and decline (< 1 cm) in CMJ height between the FO and PL group, respectively, may not have a meaningful impact on sport performance. In elite rugby players, Black et al.³⁹ reported that LC n-3 supplementation ($1.1\text{g}\cdot\text{d}^{-1}$ EPA+DHA) for 5 weeks improved CMJ performance by 4.6%, whereas CMJ decreased by 3.4% in the PL group. It is unclear why the magnitude of improvement and decline in the FO and PL group, respectively, was much greater compared to our study. This result may be partially explained by the mixed group of athletes in our sample experiencing different training routines compared to rugby players in a homogeneous training environment.

For in-season athletes, we also demonstrated that FO supplementation led to a 0.7 kg increase in LBM and a 0.1 kg loss of FM, whereas LBM and FM increased by 0.3 kg and 0.4 kg in the PL group, respectively. While the results were not significant, the body composition changes observed in the FO group were remarkably similar to those reported in previous studies.^{31,32,40–42} In regards to LBM, two studies in young, healthy adults have reported increases in LBM ranging from 0.2 kg to 0.5 kg with LC n-3 supplementation.^{40,42} Utilizing a similar LC n-3 dose as our study ($2.4\text{g}\cdot\text{d}^{-1}$ EPA+DHA), Noreen et al.⁴⁰ reported that FO significantly increased LBM by 0.5 kg over 6 weeks. While Couet et al.⁴² reported an 0.2 kg increase in LBM, the results were not significantly different than the control condition. One plausible explanation for the magnitude of effect differences may be related to the LC n-3 dose provided. Our study and that of Noreen et al.⁴⁰ supplied $> 2\text{g}\cdot\text{d}^{-1}$ LC n-3, whereas Couet et al.⁴² only provided $1.8\text{g}\cdot\text{d}^{-1}$ LC n-3 over three weeks. Evidence in clinical populations tend to agree with the implication that $\geq 2\text{g}$ LC n-3 per day may be needed to elicit a meaningful increase in LBM.^{31,32} Both trials also reported significant decreases in FM between the FO and PL groups with treatment effects of 0.58 and 0.7 kg.^{40,42} Although our study reported similar results, the between group difference was less than previously reported (0.5 kg). While the data appear to be directionally consistent with other studies in young, healthy adults, our investigation was unable to definitively show a beneficial effect of FO supplementation on body composition.

Our study was not without limitations that should be considered when interpreting our results. First, we did not track physical activity during the intervention; however, athletes participated in their regular training regimes and were all in the same phase of training (in-season). As the athletes were recruited from various sports, some of the differences between outcomes could be influenced by the markedly different training regimes based on sport. Due to contact precautions established to protect the safety of the athletes during the pandemic, the DXA protocol was

unstandardized and there was incomplete data collection. Lastly, our sample size did not allow us to compare or assess differences in outcome measures between sports. While our findings may be generalized to athletes, individual recommendations based on sport are much more tenuous.

Conclusions

In the context of in-season training, 8-weeks of LC n-3 supplementation (2.9g·d⁻¹) increases HGS in collegiate athletes and the degree of improvement is positively correlated with the increase in whole blood EPA and EPA:AA. Although we reported favorable outcomes for power and body composition in athletes taking a FO supplement, the results were less conclusive and must be interpreted with caution. Given the recently reported depletion of EPA and DHA status during physical training and the sub-optimal LC n-3 status in athletes, fish oil supplementation sustained over time has the potential to positively impact athletic performance.

Acknowledgements

The authors would like to thank the wellness and athletic staff and athletes for their willingness to participate in the study. Additionally, we'd like to thank CAS BioSciences for their generous donation of the fish oil and placebo supplements.

References

1. Oliver JM, Anzalone AJ, Turner SM. Protection before impact: the potential neuroprotective role of nutritional supplementation in sports-related head trauma. *Sports Med.* 2018;48(Suppl 1):39-52. doi:10.1007/s40279-017-0847-3
2. Siscovick D.S., Barringer T.A., Fretts A.M., et al. Omega-3 Polyunsaturated Fatty Acid (Fish Oil) Supplementation and the Prevention of Clinical Cardiovascular Disease: A Science Advisory from the American Heart Association. *Circulation.* 2017;135(15):e867-e884. doi:10.1161/CIR.0000000000000482
3. Philpott JD, Witard OC, Galloway SDR. Applications of omega-3 polyunsaturated fatty acid supplementation for sport performance. *Res Sports Med.* 2019;27(2):219-237.
4. Lewis NA, Daniels D, Calder PC, Castell LM, Pedlar CR. Are there benefits from the use of fish oil supplements in athletes? A systematic review. *Adv Nutr.* 2020;11(5):1300-1314. doi:10.1093/advances/nmaa050
5. Heileson JL, Funderburk LK. The effect of fish oil supplementation on the promotion and preservation of lean body mass, strength, and recovery from physiological stress in young, healthy adults: a systematic review. *Nutr Rev.* 2020;78(12):1001-1014.
6. McGlory C, Calder PC, Nunes EA. The influence of omega-3 fatty acids on skeletal muscle protein turnover in health, disuse, and disease. *Front Nutr.* 2019;6:144.
7. McGlory C., Galloway S.D.R., Hamilton D.L., et al. Temporal changes in human skeletal muscle and blood lipid composition with fish oil supplementation. *Prostaglandins Leukotrienes Essent Fatty Acids.* 2014;90(6):199-206.
8. Borkman M, Storlien LH, Pan DA, Jenkins AB, Chisholm DJ, Campbell LV. The relation between insulin sensitivity and the fatty-acid composition of skeletal-muscle phospholipids. *N Engl J Med.* 1993;328(4):238-244. doi:10.1056/NEJM199301283280404
9. Smith GI, Atherton P, Reeds DN, et al. Omega-3 polyunsaturated fatty acids augment the muscle protein anabolic response to hyperinsulinaemia–hyperaminoacidaemia in healthy young and middle-aged men and women. *Clin Sci.* 2011;121(6):267-278.
10. Oliver JM, Jones MT, Kirk KM, et al. Effect of docosahexaenoic acid on a biomarker of head trauma in American football. *Med Sci Sports Exerc.* 2016;48(6):974-982. doi:10.1249/MSS.0000000000000875
11. Heileson JL, Anzalone AJ, Carbuhn AF, et al. The effect of omega-3 fatty acids on a biomarker of head trauma in NCAA football athletes: a multi-site, non-randomized study. *J Int Soc Sports Nutr.* 2021;18(1):65. doi:10.1186/s12970-021-00461-1
12. Yates A, Norwig J, Maroon JC, et al. Evaluation of lipid profiles and the use of omega-3 essential Fatty Acid in professional football players. *Sports Health.* 2009;1(1):21-30. doi:10.1177/1941738108326978
13. Ritz PP, Rogers MB, Zabinsky JS, et al. Dietary and biological assessment of the omega-3 status of collegiate athletes: a cross-sectional analysis. *PLoS ONE.* 2020;15(4):e0228834.
14. Anzalone A, Carbuhn A, Jones L, et al. The omega-3 index in National Collegiate Athletic Association Division I collegiate football athletes. *J Athl Train.* 2019;54(1):7-11.
15. Davis JK, Freese EC, Wolfe AS, Basham SA, Stein KMW. Evaluation of Omega-3 Status in Professional Basketball Players. *J Strength Cond Res.* 2021;35(7):1794-1799. doi:10.1519/JSC.0000000000004023

16. Heileson JL, Elliott A, Buzzard JA, et al. A Cross-Sectional Analysis of Whole Blood Long-Chain ω -3 Polyunsaturated Fatty Acids and Its Relationship with Dietary Intake, Body Composition, and Measures of Strength and Power in Collegiate Athletes. *J Am Coll Nutr*. 2021;1-7. doi:10.1080/07315724.2021.1995910
17. Walker AJ, McFadden BA, Sanders DJ, Rabideau MM, Hofacker ML, Arent SM. Biomarker Response to a Competitive Season in Division I Female Soccer Players. *J Strength Cond Res*. 2019;33(10):2622-2628. doi:10.1519/JSC.0000000000003264
18. Huggins RA, Fortunati AR, Curtis RM, et al. Monitoring Blood Biomarkers and Training Load Throughout a Collegiate Soccer Season. *J Strength Cond Res*. 2019;33(11):3065-3077. doi:10.1519/JSC.0000000000002622
19. Davinelli S, Corbi G, Righetti S, et al. Relationship Between Distance Run Per Week, Omega-3 Index, and Arachidonic Acid (AA)/Eicosapentaenoic Acid (EPA) Ratio: An Observational Retrospective Study in Non-elite Runners. *Front Physiol*. 2019;10. doi:10.3389/fphys.2019.00487
20. Santos VC, Levada-Pires AC, Alves SR, Pithon-Curi TC, Curi R, Cury-Boaventura MF. Effects of DHA-rich fish oil supplementation on lymphocyte function before and after a marathon race. *International journal of sport nutrition and exercise metabolism*. 2013;23(2):161-169.
21. Nelson JR, Raskin S. The eicosapentaenoic acid:arachidonic acid ratio and its clinical utility in cardiovascular disease. *Postgrad Med*. 2019;131(4):268-277. doi:10.1080/00325481.2019.1607414
22. Ochi E, Yanagimoto K, Tsuchiya Y. Plasma Eicosapentaenoic Acid Is Associated with Muscle Strength and Muscle Damage after Strenuous Exercise. *Sports (Basel)*. 2021;9(1). doi:10.3390/sports9010011
23. Teixeira V, Voci SM, Mendes-Netto RS, da Silva DG. The relative validity of a food record using the smartphone application MyFitnessPal. *Nutr Diet*. 2018;75(2):219-225. doi:10.1111/1747-0080.12401
24. Sublette ME, Segal-Isaacson CJ, Cooper TB, et al. Validation of a food frequency questionnaire to assess intake of n-3 polyunsaturated fatty acids in subjects with and without major depressive disorder. *J Am Diet Assoc*. 2011;111(1):117-123.e1-2. doi:10.1016/j.jada.2010.10.007
25. Kamolrat T, Gray SR. The effect of eicosapentaenoic and docosahexaenoic acid on protein synthesis and breakdown in murine C2C12 myotubes. *Biochem Biophys Res Commun*. 2013;432(4):593-598.
26. VanItallie TB, Yang MU, Heymsfield SB, Funk RC, Boileau RA. Height-normalized indices of the body's fat-free mass and fat mass: potentially useful indicators of nutritional status. *Am J Clin Nutr*. 1990;52(6):953-959. doi:10.1093/ajcn/52.6.953
27. Sayers SP, Harackiewicz DV, Harman EA, Frykman PN, Rosenstein MT. Cross-validation of three jump power equations. *Med Sci Sports Exerc*. 1999;31(4):572-577. doi:10.1097/00005768-199904000-00013
28. Harman EA, Rosenstein MT, Frykman PN, Rosenstein RM, Kraemer WJ. Estimation of Human Power Output from Vertical Jump. *J Strength Cond Res* 1991;5(3):116-120.
29. Cohen J. *Statistical Power Analysis for the Behavioral Sciences*. 2nd ed. Lawrence Erlbaum; 1998.
30. Lee SR, Jo E, Khamoui AV. Chronic fish oil consumption with resistance training improves grip strength, physical function, and blood pressure in community-dwelling older adults. *Sports*. 2019;7(7):167. doi:10.3390/sports7070167
31. Smith GI, Julliand S, Reeds DN, Sinacore DR, Klein S, Mittendorfer B. Fish oil-derived n-3 PUFA therapy increases muscle mass and function in healthy older adults. *Am J Clin Nutr*. 2015;102(1):115-122.
32. Logan S.L., Spriet L.L. Omega-3 fatty acid supplementation for 12 weeks increases resting and exercise metabolic rate in healthy community- dwelling older females. *PLoS ONE*. 2015;10(12).
33. Krzyżmińska-Siemaszkó R, Czepulis N, Lewandowicz M, et al. The effect of a 12-week omega-3 supplementation on body composition, muscle strength and physical performance in elderly individuals with decreased muscle mass. *Int J Environ Res Public Health*. 2015;12(9):10558-10574. doi:10.3390/ijerph120910558
34. Daďová K, Petr M, Štefl M, et al. Effect of calanus oil supplementation and 16 week exercise program on selected fitness parameters in older women. *Nutrients*. 2020;12(2). doi:10.3390/nu12020481
35. Gravina L, Brown FF, Alexander L, et al. n-3 Fatty Acid Supplementation During 4 Weeks of Training Leads to Improved Anaerobic Endurance Capacity, but not Maximal Strength, Speed, or Power in Soccer Players. *Int J Sport Nutr Exer Metabol*. 2017;27(4):305-313. doi:10.1123/ijnsnem.2016-0325
36. Da Boit M, Sibson R, Sivasubramaniam S, et al. Sex differences in the effect of fish-oil supplementation on the adaptive response to resistance exercise training in older people: a randomized controlled trial. *Am J Clin Nutr*. 2017;105(1):151-158. doi:10.3945/ajcn.116.140780
37. Dalle S, Van Roie E, Hiroux C, et al. Omega-3 Supplementation Improves Isometric Strength But Not Muscle Anabolic and Catabolic Signaling in Response to Resistance Exercise in Healthy Older Adults. *J Gerontol A Biol Sci Med Sci*. 2021;76(3):406-414. doi:10.1093/gerona/glaa309

38. Crestani DM, Bonin ÉFR, Barbieri RA, Zagatto AM, Higino WP, Milioni F. Chronic supplementation of omega-3 can improve body composition and maximal strength, but does not change the resistance to neuromuscular fatigue. *Sport Sci Health*. 2017;13(2):259-265. doi:10.1007/s11332-016-0322-9
39. Black K.E., Witard O.C., Baker D., et al. Adding omega-3 fatty acids to a protein-based supplement during pre-season training results in reduced muscle soreness and the better maintenance of explosive power in professional Rugby Union players. *Eur J Sport Sci*. 2018;18(10):1357-1367.
40. Noreen EE, Sass MJ, Crowe ML, Pabon VA, Brandauer J, Averill LK. Effects of supplemental fish oil on resting metabolic rate, body composition, and salivary cortisol in healthy adults. *J Int Soc Sports Nutr*. 2010;7(1):31.
41. Hill AM, Buckley JD, Murphy KJ, Howe PRC. Combining fish-oil supplements with regular aerobic exercise improves body composition and cardiovascular disease risk factors. *Am J Clin Nutr*. 2007;85(5):1267-1274.
42. Couet C., Delarue J., Ritz P., Antoine J-M., Lamisse F. Effect of dietary fish oil on body fat mass and basal fat oxidation in healthy adults. *Int J Obes (Lond)*. 1997;21(8):637-643.