Applications of omega-3 polyunsaturated fatty acid supplementation for sport performance

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Abstract

Omega-3 (n-3) polyunsaturated fatty acid (PUFA) supplementation has recently been proposed as an ergogenic aid for athletes. This claim is primarily based on mechanistic evidence that n-3PUFA's exert anti-inflammatory properties and act to change the functional capacity of the muscle cell by modifying the membrane fluidity of proteins and lipids within the cell membrane. In this review, we critically evaluate the scientific literature that examines the efficacy of n-3PUFA supplementation to improve athlete performance within the context of promoting muscle adaptation, energy metabolism, muscle recovery and injury prevention (e.g. muscle loss during immobilisation, concussion). These findings have applications to athletes competing in strength/power-, endurance- and team-, based sports. Based on available information, there is promising scientific evidence that n-3PUFA supplementation may improve endurance capacity by reducing the oxygen cost of exercise. Moreover, several studies report a benefit of n-3PUFA supplementation in promoting recovery from eccentric-based muscle damaging exercise. In contrast, there is insufficient evidence from studies in athletic populations to support the claim that n-3PUFA supplementation facilitates muscle growth during resistance training or preserves muscle mass during catabolic scenarios such as energy restriction or immobilisation. Moving forward, there remains ample scope to investigate context-specific applications of n-3PUFA supplementation for sport performance.

Introduction

Long chain n-3 polyunsaturated fatty acids (n-3PUFA) continue to receive considerable research attention as a potential ergogenic aid in the context of enhancing sport performance. Fish oil primarily consists of the n-3PUFA's eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), whilst another n-3PUFA, α-linolenic acid (ALA), is derived from plant oils such as flaxseed and soybean oil. ALA is an 18 carbon chain fatty acid with 3 double bonds (18:3), whereas EPA is a 20 carbon chain fatty acid with 5 double bonds (20:5) and DHA is a 22 carbon chain fatty acid with 6 double bonds (22:6). Fatty acids comprise of a hydrocarbon chain with a methyl group and carboxyl group at opposing ends. Humans do not possess an enzyme called omega-3 desaturase that initiates the addition of another double bond to the 15th carbon chain. Therefore, n-3PUFA are classified as essential fatty acids because they must be provided in the diet to initiate the formation of EPA and DHA. The conversion of EPA and DHA from ALA occurs via several reaction steps. However, the complete conversion of ALA to DHA is less than 3% in males and 10% in females. This inefficient conversion rate is partly attributed to the production of omega-6 PUFA since there is competition for the desaturase and elongase phase and the typical western diet contains a higher intake of omega-6 fatty acids than omega-3 fatty acids (Burdge, Jones and Wootton, 2002; Burdge and Wootton, 2002).

The most common dietary source of n-3PUFA is oily fish. Mackerel contains approximately 3.2g of n-3PUFA per 100g serving and is considered the fish type most rich in n-3PUFA (Sprague, Dick & Tocher, 2016) **(Table 1)**. Other types of fish that contain an abundance of n-3PUFA include salmon, sardines and tuna. Alternative fish sources of n-3PUFA include walnuts, chia seeds and egg yolks. Indeed, nuts and seeds are common sources of n-3PUFA consumed by vegan athletes in order to meet daily n-3PUFA needs (Rogerson, 2017). Currently, the optimal dose of n-3PUFA for athletes or the general population is not definitely known. However, the World Health Organization indicate that individuals should

aim to consume 1-2 servings of oily fish per week, equivalent to 200-500mg of n-3PUFA per day (World Health Organisation, 2003). In comparison, the recent (2003-2008) National Health and Nutrition Health Survey revealed an average EPA and DHA intake of ~200 mg/day (Papanikolaou et al., 2014). Taken together, these data suggest that most individuals fail to meet daily n-3PUFA intake guidelines, however it is unclear whether athletes currently meet these guidelines.

It is estimated that 85% of elite athletes use at least one dietary supplement as a potential ergogenic aid (Maughan, Depiesse and Geyer, 2007). Of these supplements, n-3PUFA is one of the most popular (Shaw, Slater and Burke, 2016). Dietary n-3PUFA supplementation has been proposed to be advantageous for athletes mainly due to its anti-inflammatory properties (Li et al., 2005). Dietary n-3PUFA supplementation has been shown to inhibit the cyclooxygenase-2 (COX-2) pathway (Lim et al., 2009;) which is known to stimulate inflammation. The incorporation of n-3PUFA's into cell membranes with supplementation also alters cell membrane fluidity (Calder et al., 1994), thus modifying protein activities and cell function (Murphy, 1990). Taken together, this mechanistic information suggests that n-3PUFA supplementation has the potential to play a role in improving training adaptation, exercise recovery, and subsequent performance across athlete populations, including strength-, endurance- and team-based sport athletes.

Table 1 – Omega-3 (n-3) polyunsaturated fatty acid **content of various commonly consumed food sources**

Food Type	g EPA + DHA per 100g serving	Typical Serving Size	Number of servings to equal 1 mackerel fillet
Mackerel	3.2	81g (1 fillet)	-
Sardines	1.9	130g (1 fillet)	1.1
Farmed Salmon	1.4	94g (1 fillet)	2
Wild Salmon	0.7	94g (1 fillet)	3.9
Canned Tuna	0.2	112g (1 can drained)	11.6
Cod Loin	0.2	140g (1 loin)	9.3
Tuna Steak	0.1	120g (1 steak)	21.6
Macroalgue Fed Lamb	0.05	125g	41.5
Chicken	0.02	145g (1 breast)	89.4
Lamb	0.01	125g	207.4
Pork	0.01	100g (1 loin)	259.2
Beef	0.01	125g	207.4

DHA = Docosahexaenoic acid; EPA = Eicosapentaenoic acid. Adapted from Sprague, Dick & Tocher (2016)

The potential health benefits of n-3PUFA's were originally based on findings from epidemiological studies. The dietary intake of Inuit's living in Greenland was rich in oily fish, with ~40% of their diet consisting of fat, and ~20% of PUFA (both n-3 and n-6) (Bang, Dyerberg and Sinclair, 1980). In comparison to a Danish diet, Inuits ingested higher amounts of n-3PUFA and reported a lower incidence rate of cardiovascular disease (CVD). The high fat content of the Inuit's diet and lower incidence of CVD was attributed to the high proportion of n-3PUFA in their diet. More recent research suggests that n-3PUFA supplementation may lower risk factors associated with CVD (Zucker et al., 1988; Bhathena et al., 1991). However, findings from meta-analyses reveal that the effects of n-3PUFA supplementation on CVD are unclear (Hooper et al., 2006; Marik and Varon, 2009; Djoussé et al., 2012). Given these health benefits, recently there has been growing interest in the application of n-3PUFA in the context of athlete performance, specifically in the context of training adaptation, exercise recovery, injury prevention and illness.

Strength/power-based Athletes

Previous research has investigated the influence of n-3PUFA supplementation on acute measurements of muscle protein synthesis (MPS) and chronic measurements of changes in muscle mass and neuromuscular function. This line of research is based on the idea that n-3PUFA ingestion sensitises skeletal muscle to the main anabolic stimuli, namely resistance exercise training and protein ingestion. The primary metabolic driver of muscle hypertrophy is an increased stimulation of MPS in response to exercise and nutrition (Biolo et al., 1997). Early proof-of-concept studies demonstrated that dietary n-3PUFA supplementation potentiated the response of MPS to amino acid provision, administered intravenously as a hyperaminoacidemic/hyperinsulinemic clamp (Smith et al., 2011a; Smith et al., 2011b) in

young (Smith et al., 2011b) and older (Smith et al., 2011a) adults. Although no changes in basal rates of MPS were observed following 8 weeks of n-3PUFA supplementation, postprandial rates of MPS and the phosphorylation status of anabolic signalling proteins within the mechanistic target of rapamycin complex (mTORC) pathway were potentiated after n-3PUFA supplementation. Consistent with this observation, research from our laboratory demonstrated an increase in skeletal muscle omega-3 lipid content and stimulation of focal adhesion kinase (FAK) —a key signalling protein that regulates MPS— following four weeks of 5g/day n-3PUFA supplementation in active males (McGlory et al., 2014). The incorporation of n-3PUFA into a muscle cell membrane has been shown to alter the cell's integrity, disrupting the fluidity of proteins and lipids within the cell membrane (Calder et al., 1994). Such structural changes in membrane composition have been proposed to provide a mechanistic explanation for improvements in cell function with n-3PUFA ingestion (Murphy, 1990).

Our research also suggests that a minimum supplementation period of 2 weeks is required to observe an increased incorporation of n-3PUFA into the muscle cell (McGlory et al., 2014), or specifically into the cell membrane. Whereas the incorporation of n-3PUFA into the muscle cell continued to increase after 4 weeks of supplementation, no plateau was observed in this study. These data suggest that > 4 weeks of n-3PUFA supplementation is required to maximise muscle incorporation of n-3PUFA. However, a systematic study is warranted to confirm this assertion. As a note of caution, it also should be highlighted that n-3PUFA muscle cell concentration, rather than muscle cell membrane concentration was measured in this study (McGlory et al., 2014). Therefore, it is assumed that the incorporation of n-3PUFA into the muscle cell also translated to the membrane.

There is mechanistic evidence from in vitro studies using muscle cell lines that EPA, rather than DHA, is the primary anabolic component of n-3PUFA (Kamolrat and Gray, 2013). In this study, the incubation of C_2C_{12} myotubules with EPA resulted in increased rates of MPS

and decreased rates of muscle protein breakdown (MPB). In contrast, incubation with DHA elicited no changes in MPS or MPB. Utilising a physiologically relevant research design, we recently investigated the influence of 8 weeks of n-3PUFA supplementation (5g/day) on the response of MPS to ingesting 30 g of whey protein with and without resistance exercise in resistance-trained young men (McGlory, et al., 2016). In contrast to previous proof-of-concept studies (Smith et al., 2011a; Smith et al., 2011b), no differences in MPS and anabolic signalling were observed between n-3PUFA and placebo (coconut oil) conditions. Our previous research suggests that ~20g of whey protein stimulates a maximal response of MPS following leg-only resistance exercise (Witard et al., 2014). Thus, it is conceivable that the 30g dose of whey protein administered in McGlory et al., (2016) saturated the muscle protein synthetic machinery, meaning that n-3PUFA supplementation could then not provide an additional stimulus for MPS. Therefore, further research is needed to investigate whether the addition of n-3PUFA to a sub-optimal dose of protein would further stimulate MPS following resistance exercise compared to a protein dose alone.

While MPS is the gold standard acute marker of muscle growth, a handful of chronic intervention studies have directly measured changes in muscle growth or strength in response to a period of n-3PUFA supplementation. In a recent study, older adults underwent 6 months of either n-3PUFA (3.36 g/day EPA + DHA) or corn oil supplementation (Smith et al., 2015). Thigh muscle volume, handgrip strength and 1-RM strength all increased in the n-3PUFA group, where no changes were detected in placebo. However, there were no differences in body mass or body fat between the two conditions. Interestingly, only the thigh was measured for muscle volume. Given that thigh muscle volume increased it was assumed that muscle mass was increased at the whole body level. Interestingly, no studies have measured the response of muscle growth or strength in response to n-3PUFA supplementation in young adults or athletic populations. Further studies should be designed to examine the responsiveness of muscle

strength and volume to a chronic (12 wk- 6 mo) period of n-3PUFA supplementation in power/strength-based athletes.

The first study to measure muscle strength following a period of n-3PUFA supplementation observed an increase in peak torque with 90 or 150 days of n-3PUFA supplementation at a dose of 2g/day (Rodacki et al., 2012). Similarly, training-induced improvements in neuromuscular function, such as muscle activation and electromechanical delay in various muscles including the bicep femoris and vastus lateralis, were enhanced with n-3PUFA vs the training only group (Rodacki et al., 2012). Since DHA is abundant within brain neurons (Kim, Huang and Spector, 2014), improvements in neuronal adaptation with n-3PUFA supplementation may indicate that neural pathways are modified. However, given that participants in this study were older females, caution should be applied when interpreting these results for athletes. Taken together, these data support a potential anabolic role for n-3PUFA ingestion in the context of preserving muscle mass in older adult populations. However, based on current information, there is limited information available to support an anabolic role of n-3PUFA for muscle growth in athletes.

Endurance-based Athletes

An important aspect of endurance exercise performance and training adaptation is the capacity to utilize substrates efficiently and maximise available energy from adenosine triphosphate (ATP) stores. The mitochondrial content of the cell aids regulation of ATP resynthesis. A key regulator of mitochondrial biogenesis, defined as the process of increasing mitochondrial volume, is Peroxisome proliferator-activated receptor-gamma coactivator (PGC-1α). Previous studies in rodents have shown dietary n-3PUFA supplementation to increase expression of PGC-1α (Hancock et al., 2008) and increase mitochondrial biogenesis

(Turner et al., 2007). However, studies investigating n-3PUFA supplementation and mitochondrial biogenesis in humans are limited. Currently, only one study has examined mitochondrial biogenesis with n-3PUFA supplementation and reported that EPA supplementation stimulated mitochondrial biogenesis in obese individuals (Laiglesia et al., 2016). Based on findings from animal studies, it is possible that n-3PUFA supplementation may increase mitochondrial biogenesis leading to improved endurance performance as mediated via the PGC-1α pathway. However, human studies in athletes are warranted in order to examine this theory.

Dietary intake of n-3PUFA also are known to alter membrane fatty acid composition in skeletal (Andersson et al., 2002) and myocardial (Charnock et al., 1992) muscle tissue. These changes in membrane composition can lead to changes in insulin sensitivity (Borkman et al., 1993) via a yet to be determined mechanism. However, a pre-clinical rodent study demonstrated that the addition of n-3PUFA to a high fat diet increased the protein expression of Glucose transporter type-4 (GLUT4; Lanza et al., 2013). GLUT4 is present only in skeletal muscle and adipose tissue and plays a key role in transporting extracellular glucose into cells that are insulin sensitive (Huang and Czech, 2007). In humans, n-3PUFA supplementation also has been shown to improve insulin sensitivity in skeletal muscle (Borkman et al., 1989). Thus, in theory, an increase in GLUT4 expression with n-3PUFA supplementation may play a key role in improving tissue insulin sensitivity and thus endurance performance.

Dietary n-3PUFA supplementation also has been shown to reduce oxygen consumption (Peoples et al., 2008; Kawabata et al., 2014), heart rate (Peoples et al., 2008) and perceived exertion (Kawabata et al., 2014) during endurance exercise. The mechanism that underpins the improved oxygen efficiency with n-3PUFA supplementation is unclear, and paradoxically n-3PUFA supplementation has been shown to initiate an increase in resting metabolic rate (Logan and Spriet, 2015). Although speculative, the increase in resting metabolic rate with n-3PUFA

ingestion may primarily be due to the increased incorporation of DHA into the cell membrane that has been shown to lead to an increase in Ca^{2+} ATPase and Na^+/K^+ ATPase activity that requires more ATP utilization (Hulbert et al., 2005). A potential mechanism that may underpin an alteration in the oxygen cost of exercise is through an increase in insulin sensitivity. Intuitively, an increase in insulin sensitivity leads to greater muscle glycogen resynthesis and the subsequent potential to increase carbohydrate oxidation rates and decrease fat oxidation rates (Watt et al., 2002). During endurance exercise, a shift in substrate utilization from fat to carbohydrate would reduce the volume of oxygen used to meet demands for ATP resynthesis, and in turn improve the calculated exercise efficiency (Cole et al., 2014).

At present, a limited number of studies have examined the influence of n-3PUFA ingestion on markers of energy metabolism and performance in endurance-trained individuals **(Table 2)**. In trained cyclists with low habitual n-3PUFA intake, eight weeks of high or low dose DHA-rich n-3PUFA supplementation resulted in a reduced oxygen cost during a cycling time trial compared to a soy bean placebo condition (Hingley et al., 2017). However, the observed increase in omega-3 index and reduction in oxygen cost did not translate into a performance advantage, with no improvements in time trial completion time, mean power during the time trial and quadriceps strength. Further research in endurance athletes is warranted to examine the impact of n-3PUFA supplementation on oxygen kinetics during exercise when oxygen availability is limited, e.g. competition and training at high altitude.

Previous research also has shown that n-3PUFA supplementation has the potential to lower heart rate and blood pressure during exercise. In elite Australian Rules footballers, 5 weeks of DHA rich n-3PUFA (1.56 g/day DHA and 0.36g/day EPA) supplementation significantly lowered heart rate during steady state submaximal exercise, however peak heart rate did not change compared to a sunflower oil placebo condition (Buckley et al., 2008). Interestingly, diastolic blood pressure increased after 5 weeks of sunflower oil supplementation

but did not change in the n-3PUFA group. Previous research demonstrates that DHA rather than EPA is the active lipid component of n-3PUFA's in reducing blood pressure and heart rate in humans (Mori et al., 1999). As a logical follow study, healthy males demonstrated a reduction in heart rate during a bout of steady state cycling with DHA rich n-3PUFA supplementation (Macartney et al., 2014). However, during repeated sprints there were no differences in heart rate between conditions. Taken together, these data suggest that whereas the provision of DHA rich n-3PUFAs results in a decreased heart rate response during submaximal exercise, at higher exercise intensities n-3PUFA supplementation has no impact on the heart rate response. The mechanism responsible for the modulation of heart rate by n-3PUFA supplementation is thought to involve multiple physiological processes including the regulation of systolic and diastolic left ventricular function, sympathetic activity and vagal tone (Mozaffarian, Gottdiener & Siscovick, 2006; O'Keefe et al., 2006). For instance, n-3PUFA supplementation is known to increase stroke volume by increasing the amount of blood ejected from the heart with each contraction, this increase in stroke volume results in a decrease in heart rate. However, further research is needed in order to fully understand the mechanisms by which n-3PUFA supplementation lowers heart rate.

Although not universally accepted, endurance athletes are often considered more susceptible to developing an upper respiratory tract infection (URTI) that can disrupt training and competitive performance (Peters & Bateman, 1983). Dietary n-3PUFA supplementation has been shown to upregulate the signalling network between cells involved in immune function, resulting in the stimulation of CD4 and CD8 lymphocyte production, thus improving the ability of immune cells to destroy foreign pathogens (de Lourdes Nahhas Rodacki et al., 2015). In this regard, a recent study examined the influence of adding n-3PUFA to other nutrients (1.1 g/day of n-3PUFA, 10 μg/day Vitamin D and 8 g/day of whey protein isolate) vs. a carbohydrate placebo control on markers of immune function in young active males and

females that continued their habitual training over a 16 week period (Da Boit et al., 2015). Although no differences in markers of immune function were observed between groups, the frequency and duration of URTI symptoms was reduced in the n-3PUFA group. However, it should be noted that diagnosis of URTIs was self reported and not clinically diagnosed by a doctor. Moreover, based on these findings alone, it is impossible to differentiate between the effects of n-3PUFA, vitamin D, whey protein or the combination of all the ingredients in observed reduction in symptoms days.

Table 2 –Studies investigating the influence of omega-3 (n-3) polyunsaturated fatty acid **supplementation on endurance capacity and performance**

Authors	Participants	Supplement Dose	Supplementation Period	Exercise	Observations
Bortolotti *et al.* (2007)	Sedentary males (n=8)	7.2g/d FO vs. PLA	14 days prior to exercise	30 min cycling (50% VO_{2max})	→ energy efficiency → VO_{2max}
Buckley *et al.* (2009)	Elite Australian Rules League Footballers (n=25)	1.92g/d n-3PUFA vs. SO	5 weeks prior to exercise	Steady-state submaximal running	↓ heart rate ↓ blood pressure → performance → recovery
Hingley *et al.* (2017)	Trained cyclists + runners (n=26)	700mg/d n-3PUFA vs. SBO	8 weeks prior to exercise	Cycling sprints 5 min time trial	↓ oxygen cost → performance
Kawabata *et al.* (2014)	Recreational team-sport males (n =20)	3.6g/day FO vs. PLA	8 weeks prior to exercise	VO_{2max} test and steady state cycling tests	→ VO_{2max} ↓ Oxygen consumption ↓ Rate of perceived exertion
Macartney et al (2014)	Healthy males (n=39)	700mg/d n-3PUFA vs. SBO	8 weeks prior to exercise	Maximal cycling sprints and 5 min time trial	↓ Submaximal and recovery heart rate → Peak heart rate
Ninio *et al* (2008)	Sedentary overweight adults (n=75)	1.92g/day n-3PUFA vs. SO	12 weeks prior to exercise	Graded submaximal test	↓ Resting and submaximal heart rate ↑ heart rate variability
Oostenbrug *et al* (1997)	Trained cyclists (n=24)	6g/d FO + Vitamin E vs. 6 g/g FO vs. PLA	3 weeks prior to exercise	Wmax and endurance cycling tests	→ time to exhaustion → VO_{2max} → maximal power
Peoples *et al* (2008)	Trained cyclists (n=20)	3.2 g/d n-3PUFA vs. OO	8 weeks prior to exercise	Submaximal exercise tests (55% of peak workload)	↓ Peak and submaximal heart rate ↓ Oxygen consumption
Rontoyanni *et al* (2012)	Healthy males (n=22)	4.7 g/d DHA vs. 4.7 g/d EPA vs. SO	Single dose	12 min multi-stage test	→ cardiac output ↓ Systemic vascular resistance
Zebrowska *et al* (2015)	Trained cyclists (n=13)	1.3g/d n-3PUFA vs. PLA	3 weeks prior to exercise	VO_{2max} test	↑ VO_{2max} ↑ endothelial function

n-3PUFA = omega-3 polyunsaturated fatty acid; DHA = Docosahexanoic acid; EPA = Eicosapentaenoic acid; PLA = Placebo; FO = Fish oil; OO = Olive oil; SO = Sunflower Oil; SBO = Soy Bean Oil.

Team-based Athletes

The initial 96 hours following exercise is commonly defined as the acute exercise recovery period (Pereira Panza et al., 2015). This period is considered crucial in optimising athlete performance, particularly during situations such as fixture congestion for team sport athletes. Repeated eccentric-based muscle contractions are known to cause damage to skeletal muscle fibres (Nedelec et al., 2012). Muscle damaging exercise has been shown to subsequently impair sport-specific performance (Eston et al., 1996). There is biological rationale behind the notion that n-3PUFA has the potential to promote recovery from muscle damaging exercise. In theory, n-3PUFA have the potential to protect the muscle from damaging exercise by increasing the structural integrity of the muscle cell membrane. Alternatively, n-3PUFA have the potential to accelerate the recovery process. In this regard, dietary n-3PUFA exhibit anti-inflammatory properties via several pathways. These pathways include inhibition of the COX-2 pathway (Li et al., 2005), the synthesis of lipoxins and resolvins that both exhibit anti-inflammatory functions (Janakiram, Mohammed and Rao, 2011) and also by reducing chemotaxis of neutrophils and reduce generation of leukotrienes, a family of inflammatory mediators produced by leukocytes (Lee et al., 1985). Therefore, it is intuitive that n-3PUFA supplementation could improve recovery following muscle damaging exercise either by preserving muscle membrane integrity of reducing inflammation.

A series of experimental studies have examined the influence of n-3PUFA ingestion on recovery from muscle damaging exercise and have revealed mixed results (Tsuchiya et al., 2016; Gray et al., 2014) **(Table 3).** A recent study examined the impact of acute supplementation with a high (15:1 ratio of EPA to DHA) or low (15:1 ratio of EPA to DHA) dose of n-3PUFA on exercise recovery (Jakeman et al., 2017). The authors reported that the group consuming the high dose of n-3PUFA observed an attenuated decrement in squat jump performance. However, no differences in markers of muscle soreness and putative blood

markers of muscle damage (e.g. creakine kinase (CK)) and inflammation (interleukin-6) were observed between conditions. These data suggest that the high ratio of EPA to DHA may be the key factor in helping to maintain performance following acute supplementation and muscle damaging exercise. However, it is difficult to interpret these data given that at least 2 weeks is required for incorporation of omega-3 into muscle tissue. Therefore, any physiological effect of n-3PUFA in this study must have been systemic (Jakeman et al., 2017).

Another recent study measured the impact of medium term (21 days prior to muscle damage) n-3PUFA supplementation on indices of recovery following muscle damaging exercise in females (McKinley-Barnard et al., 2018). Participants consumed either 4.2 g/day of n-3PUFA or a placebo supplement consisting of safflower oil for 21 days before undergoing a bout of intense eccentric exercise. Supplementation of n-3PUFA failed to attenuate muscle soreness and inflammation measured 24 hours following exercise compared to the placebo condition. Unfortunately, this study did not collect measurements of exercise recovery 48, 72 or 96 hours post-exercise and therefore may have missed important information regarding the effectiveness of n-3PUFA ingestion in promoting acute exercise recovery.

Whilst this body of work **(Table 3)** provides proof-of-concept for the potential role of n-3PUFA ingestion in accelerating recovery from muscle damaging exercise, the direct application of these results to team-based athletes should be considered with caution for several reasons. First, these studies are typically performed in untrained participants in which a high degree of muscle damage is likely after unaccustomed exercise. Hence, it may be argued that the application of results is more appropriate in the context of improving compliance of previously sedentary population to a new exercise routine, rather than the elite team-sport athlete with the goal of complete recovery prior to the next match. Second, the ecological validity of the muscle damage protocol used in this study is not directly relevant to sporting

movements. Finally, the sensitivity and specificity of endpoint measurements to team sport athletes is weak.

To address these limitations, we recently recruited competitive soccer players to ingest a combined n-3PUFA (2.8 g/day) and whey protein (30 g/day) supplement beverage over a 6 week period prior to performing an intense exercise bout (Philpott et al., 2018). In the 72 hours following the muscle damaging exercise, the soccer players in the n-3PUFA plus protein group reported reduced levels of muscle soreness. The n-3PUFA group also experienced a reduction in plasma creatine kinase concentrations as a putative blood marker of muscle damage, compared to the whey protein beverage only, or the carbohydrate placebo beverage. As such, these data imply that n-3PUFA supplementation protected the muscle cell from the muscle damage protocol and therefore soccer players experienced less damage during exercise. However, there was no influence of n-3PUFA ingestion on soccer performance tests such as the yoyo intermittent recovery test or the Loughborough soccer passing test, which arguably offer greater application for recovery in the team sport athlete. To better understand the impact of n-3PUFA on recovery this study needs to be replicated in a real life football situation, using a simulated soccer match.

Our recent research also has observed that four weeks of n-3PUFA supplementation in soccer players resulted in improved anaerobic endurance running capacity while maintaining their habitual training schedule (Gravina et al., 2017). Over 4 weeks of training, soccer players experienced an increase of 203m in the Yo-Yo level 1 test following ingestion of 0.1 g/kg/day of n-3PUFA, compared to only a 62 m improvement in the placebo group. However, adaptations in power, speed and maximal knee extensor strength were not influenced by the omega-3 supplementation. Therefore, it is possible that n-3PUFA supplementation may improve high intensity running capacity in soccer players, but further research is needed to investigate different athlete populations.

Table 3 – Studies investigating the influence of omega-3 (n-3) polyunsaturated fatty acid **supplementation on indices of recovery following exercise-induced muscle damage**

Authors	Participants	Supplement Dose	Supplementation Period	Muscle Function	Muscle Soreness	Muscle Damage Markers (CK, Mb etc.)
Corder *et al.* (2016)	Healthy females (n=27)	3g/d DHA vs. PLA	7 days prior and 2 days after exercise		DHA < PLA	DHA = PLA
DiLorenzo *et al.* (2014)	Untrained males (n=41)	2g/d DHA vs. PLA	28 days prior to exercise	DHA = PLA	DHA = PLA	DHA < PLA
Gray *et al.* (2014)	Males (n=20)	3g/d FO vs. 3g/d OO	6 weeks prior to exercise	FO = OO	FO = OO	FO < OO
Jakeman *et al.* (2017)	Healthy active males (n=27)	800mg/10kg/BM EPA vs. 250mg/10kg/BM EPA vs. PLA	Single dose	High EPA < Low EPA, PLA	High EPA = Low EPA, PLA	High EPA = Low EPA, PLA
Jouris *et al.* (2011)	Healthy males (n =11)	2g/day EPA + 2g/day DHA vs. PLA	7 days prior to exercise		FO < PLA	
Lembke et al (2014)	Healthy males and females (n=63)	2.7g/day FO vs. PLA	30 days prior to exercise			FO < PLA
McKinley-Barnard et al (2018)	Healthy active females (n=22)	4.2 g/day n-3PUFA vs. PLA	21 days prior to exercise		FO = PLA	FO = PLA
Philpott et al (2018)	Male soccer players (n=30)	2.2 g/day FO + PRO + CHO vs. PRO + CHO vs. CHO	42 days prior to exercise and 2 days following exercise	FO = PRO, CHO	FO < PRO, CHO	FO = PRO, CHO
Tartibian *et al* (2011)	Untrained males (n=45)	1.8g/day FO vs. PLA	30 days prior and 2 days following exercise.			FO < PLA
Tsuchiya et al (2016)	Healthy males (n=24)	2.4g/day n-3PUFA vs. CO	56 days prior and 5 days following exercise.	n-3PUFA > CO	n-3PUFA < CO	n-3PUFA < CO

DHA = Docosahexaenoic acid; EPA = Eicosapentaenoic acid; PLA = Placebo; n-3PUFA = n-3PUFA; OO = Olive oil; FO = Fish oil; CHO = Carbohydrate; PRO = Protein; CO = Corn Oil.

Special Considerations

Energy Restriction

Athletes competing in weight category sports often undergo periods of energy restriction. Periods of sustained energy restriction are often accompanied by the loss of muscle mass (Mettler, Mitchell & Tipton, 2010; Weinheimer, Sands & Campbell, 2010) due, primarily, to a reduction in basal rates of MPS (Pasiakos et al., 2010) rather than an increase in MPB (Longland et al., 2016). Within clinical studies (e.g. cancer patients), n-3PUFA supplementation has been shown to attenuate the loss of muscle mass (Murphy et al., 2011). Within an athletic setting, a recent study from our laboratory examined the influence of n-3PUFA supplementation during 2 weeks of energy restriction on lean and fat mass loss in resistance-trained athletes (Philpott et al., unpublished). Athletes underwent 2 weeks of 40% calorie restriction with the nutritional composition of 50% carbohydrate, 35% fat and 15% protein. Half of the participants (n=10) supplemented with an n-3PUFA beverage on a twice daily basis, and the other participants supplemented with a carbohydrate placebo beverage while continuing with their habitual training programme. Following the 2 weeks of supplementation, participants lost similar amounts of body mass, muscle mass and fat mass independent of which supplement beverage was consumed. While these initial data do not support the use of n-3PUFA ingestion during periods of energy restriction, future studies should examine the effects of n-3PUFA supplementation on the attenuation of muscle mass over a longer period of energy restriction in athletes.

Immobilisation

Serious injury in athletes can result in limb immobilisation. The muscle atrophy associated with periods of immobilisation is due, at least in part, to an attenuated response of MPS to ingested protein (Wall 2013); a concept known as anabolic resistance. Pre-clinical

studies have used a rodent model to investigate the influence of n-3PUFA supplementation on muscle mass. This work demonstrated that rats consuming a diet consisting of 2% corn oil and 5% cod liver oil retained myosin heavy chain content and inhibited the COX-2 pathway as an inflammatory marker following a 10 day period hind-limb immobilisation compared to rats consuming a diet consisting of 7% corn oil alone (You, et al., 2010a). However, when the hind limbs of the rats were remobilized for 13 days following the 10 day hind-limb immobilization (You et al., 2010b) the fish oil group experienced an impaired recovery of myosin heavy chain content compared to the corn oil group. Moreover, following remobilisation the phosphorylation status of mTORC-associated anabolic signalling proteins were increased with corn oil compared to n-3PUFA during the early stages of remobilisation (3 days). Taken together, these data indicate that n-3PUFA ingestion is effective in retaining muscle mass during the immobilisation period. However, n-3PUFA may not influence, and even possibly inhibits, the recovery process during the remobilisation phase (You et al., 2010b). Follow up human research is warranted in order to examine the impact of n-3PUFA supplementation during periods of immobilisation and remobilisation following injury.

Concussion

The diagnosis and treatment of concussion is currently a hot topic in Sport Nutrition. DHA is abundant in the plasma membranes of the brain which is involved in neuronal signalling (Fontani et al., 2005). Early research examining the effects of n-3PUFA supplementation on recovery from concussion has been conducted primarily in rat models. One of the earliest studies elicited mild traumatic brain injury to rats before conducting the Morris Water Maze test to assess performance on consecutive days 10-14 after the traumatic brain injury (TBI) (Wang et al., 2013). Rats either consumed a diet consisting of 6% n-3PUFA or a diet of 6% soybean oil before and during the recovery phase of brain injury. Rats that consumed the n-3PUFA diet managed to complete the maze faster than the placebo group during TBI

recovery Due to ethical reasons, studying recovery from concussion in humans is challenging. However, a recent study did examine the effect of n-3PUFA supplementation over a full season in American football players. These data revealed n-3PUFA ingestion (2, 4 or 6 g/day) decreased concentrations of serum neurofilament light, a biomarker of head trauma (Oliver et al., 2016). However, more research is required to determine the effectiveness of n-3PUFA in the treatment of TBI and concussion in contact sport athletes.

Bleeding

EPA is known to replace arachidonic acid in the phospholipid layer of platelet cell membranes following n-3PUFA ingestion (Lorenz et al., 1983). As a consequence, platelet aggregation may be reduced due to a reduction in levels of thromboxane A within the plasma. Platelets mediate the wound healing process via blood clotting. Thus, in theory a decrease in platelet aggregation may increase bleeding time. Consistent with this notion, a recent study also suggests that n-3PUFA supplementation may reduce platelet aggregation in healthy individuals and therefore increase bleeding time following surgery or lacerations (McEwen et al., 2013). However, human studies examining the influence of n-3PUFA supplementation on bleeding time and severity have generally shown mixed results. A recent systematic review found no difference in bleeding risk with n-3PUFA supplementation in different populations, including athletes (Begtrup, Krag & Hvas, 2017). However, the interpretation of this systematic review may be influenced by variations in the dose and duration of n-3PUFA supplementation between studies. Overall, although n-3PUFA supplementation may reduce platelet aggregation there appears to be no effect on bleeding rates following surgery. Therefore, unless athletes are ingesting a high dose of n-3PUFA, concerns over bruising and bleeding following an injury during sport appear unfounded.

Current issues in omega-3 and sport performance research

As with all nutritional supplements, more research is needed to examine the effects of n-3PUFA on athletic performance. Current issues regarding n-3PUFA supplementation on sport performance are four-fold. First, it is currently unknown the duration of time in which n-3PUFA concentrations in muscle and blood return to baseline after cessation of supplementation. This information would be useful in the design of crossover studies that investigate the impact of n-3PUFA ingestion on a chosen marker of sport performance.

Second, significant variation exists with regards to the dose and duration of n-3PUFA supplementation employed between studies. As discussed previously, at least two weeks of n-3PUFA supplementation is sufficient to detect an increase in n-3PUFA concentration within the muscle lipid pool, but it is not yet known how many weeks is required to maximise this response. Moreover, the optimum dose of n-3PUFA to reduce inflammation, maximise the incorporation of EPA and DHA into the muscle membrane and improve various aspects of sport performance remains unknown.

Third, research examining the effects of fish oil derived n-3PUFA on athletic performance have utilised a range of placebos, including safflower oil, corn oil and coconut oil. Corn oil and safflower oil both contain high amounts of omega-6, so when used as a placebo these oils actually alter the omega 3 to omega 6 ratio. To date, the most appropriate placebo appears to be coconut oil which does not contain any omega-6 or omega-3. However, there is evidence that the short chain saturated fats in coconut oil may have an impact on metabolism (Eyres et al., 2016). Therefore, the most appropriate fish oil placebo for all research testing has yet to be established.

Finally, the ratio of EPA to DHA present within the n-3PUFA supplement should be considered when interpreting the findings from research into the applications of n-3PUFA ingestion for sport performance. Current research has used multiple different ratios of EPA to

DHA whether that is 1:1, EPA rich or DHA rich supplementation. Given that EPA and DHA exhibit their own active properties and act independently, caution should be applied when interpreting n-3PUFA supplementation research with different ratios of EPA to DHA.

Practical applications and conclusion

The applications of n-3PUFA supplementation for sport performance are relevant to athletes from strength, endurance and team based sports, with recommendations tailored to the specific performance goals of the athlete (table 4). Based on currently available scientific evidence, there is potential for n-3PUFA supplementation to improve muscle adaptation, energy metabolism, muscle recovery and injury prevention. As such, n-3PUFA supplementation for athletes may yet prove to be effective, and at the very least not detrimental, for performance, except potentially following immobilisation. However, more research is needed to further investigate some of the promising applications of n-3PUFA supplementation on skeletal muscle mass retention, and growth, as well as in recovery from concussion in athletic populations.

Table 4 – Practical applications of omega-3 polyunsaturated fatty acid supplementation for athletic performance

Athlete/context	Strength of Evidence*	Practical Application
Strength/power-based Athletes	3	• Dietary n-3PUFA supplementation switches on mTORC-related signalling proteins involved in stimulating muscle protein synthesis.
	2	• However, preliminary evidence suggests that ingesting sufficient dietary protein negates any additive effect of n-3PUFA supplementation on muscle protein synthesis.
Endurance-based Athletes	3	• Dietary n-3PUFA supplementation appears to increase oxygen efficiency during endurance exercise. However, whether this physiological response translates to a performance improvement remains unclear.
	2	• Current evidence linking n-3PUFA supplementation with a reduced incidence of URTI in endurance-based athletes is only preliminary at present and warrants follow up study.
Team-based Athletes	3	• The role of dietary n-3PUFA supplementation in the day-to-day recovery of team-sport athletes for reducing muscle soreness and attenuating the decline in muscle function is promising and warrants follow up study.
	2	• The role of dietary n-3PUFA supplementation in promoting adaptation to training in team-based athletes is promising and warrants follow up study.
Energy Restriction	2	• Dietary n-3PUFA supplementation does not appear to benefit the retention of muscle mass during periods of energy restriction.
Immobilisation	2	• There is preliminary evidence, albeit in rodent models, that n-3PUFA supplementation may attenuate the loss of muscle mass during limb immobilisation.
Concussion	2	• There is preliminary evidence that n-3PUFA supplementation exerts a protective effect during concussion-related injury.
Bleeding	4	• Dietary n-3PUFA supplementation does not impact bleeding rates following minor wounds or surgery.

* Strength of evidence grading: 5 - very strong, 4 - strong, 3 - medium, 2 - weak, 1 - very weak.

n-3PUFA = omega-3 polyunsaturated fatty acid; mTORC = mechanistic target of rapamycin; URTI = upper respiratory tract infection.

Reference List

Andersson, A., Nälsén, C., Tengblad, S., & Vessby, B. (2002). Fatty acid composition of skeletal muscle reflects dietary fat composition in humans. *The American Journal of Clinical Nutrition*, *76*(6), 1222–1229.

Appell, H. J. (1990). Muscular atrophy following immobilisation. A review. *Sports Medicine (Auckland, N.Z.)*, *10*(1), 42–58.

Bang, H. O., Dyerberg, J., & Sinclair, H. M. (1980). The composition of the Eskimo food in north western Greenland. *The American Journal of Clinical Nutrition*, *33*(12), 2657–2661.

Begtrup, K. M., Krag, A. E., & Hvas, A.-M. (2017). No impact of fish oil supplements on bleeding risk: a systematic review. *Danish Medical Journal*, *64*(5).

Biolo, G., Tipton, K. D., Klein, S., & Wolfe, R. R. (1997). An abundant supply of amino acids enhances the metabolic effect of exercise on muscle protein. *American Journal of Physiology-Endocrinology and Metabolism*, *273*(1), E122–E129.

Bhathena, S. J., Berlin, E., Judd, J. T., Kim, Y. C., Law, J. S., Bhagavan, H. N., … Nair, P. P. (1991). Effects of ω3 fatty acids and vitamin E on hormones involved in carbohydrate and lipid metabolism in men. *The American Journal of Clinical Nutrition*, *54*(4), 684–688.

Borkman, M., Chisholm, D. J., Furler, S. M., Storlien, L. H., Kraegen, E. W., Simons, L. A., & Chesterman, C. N. (1989). Effects of fish oil supplementation on glucose and lipid metabolism in NIDDM. *Diabetes*, *38*(10), 1314–1319.

Borkman, M., Storlien, L. H., Pan, D. A., Jenkins, A. B., Chisholm, D. J., & Campbell, L. V. (1993). The Relation between Insulin Sensitivity and the Fatty-Acid Composition of Skeletal-Muscle Phospholipids. *New England Journal of Medicine*, *328*(4), 238–244.

Bortolotti, M., Tappy, L., & Schneiter, P. (2007). Fish oil supplementation does not alter energy efficiency in healthy males. *Clinical Nutrition*, *26*(2), 225–230.

Buckley, J. D., Burgess, S., Murphy, K. J., & Howe, P. R. C. (2009). DHA-rich fish oil lowers heart rate during submaximal exercise in elite Australian Rules footballers. *Journal of Science and Medicine in Sport*, *12*(4), 503–507.

Burdge, G. C., Jones, A. E., & Wootton, S. A. (2002). Eicosapentaenoic and docosapentaenoic acids are the principal products of α-linolenic acid metabolism in young men. *British Journal of Nutrition*, *88*(04), 355.

Burdge, G. C., & Wootton, S. A. (2002). Conversion of α-linolenic acid to eicosapentaenoic, docosapentaenoic and docosahexaenoic acids in young women. *British Journal of Nutrition*, *88*(04), 411.

Calder, P. C., Yaqoob, P., Harvey, D. J., Watts, A., & Newsholme, E. A. (1994). Incorporation of fatty acids by concanavalin A-stimulated lymphocytes and the effect on fatty acid composition and membrane fluidity. *The Biochemical Journal*, *300 (Pt 2)*(Pt 2), 509–518.

Charnock, J. S., McLennan, P. L., & Abeywardena, M. Y. (1992). Dietary modulation of lipid metabolism and mechanical performance of the heart. *Molecular and Cellular Biochemistry*, *116*(1–2), 19–25.

Cole, M., Coleman, D., Hopker, J., & Wiles, J. (2013). Improved Gross Efficiency during Long Duration Submaximal Cycling Following a Short-term High Carbohydrate Diet. *International Journal of Sports Medicine*, *35*(03), 265–269.

Corder, K. E., Newsham, K. R., McDaniel, J. L., Ezekiel, U. R., & Weiss, E. P. (2016). Effects of Short-Term Docosahexaenoic Acid Supplementation on Markers of Inflammation after Eccentric Strength Exercise in Women. *Journal of Sports Science & Medicine*, *15*(1), 176–183.

Da Boit, M., Gabriel, B., Gray, P., & Gray, S. (2015). The Effect of Fish Oil, Vitamin D and Protein on URTI Incidence in Young Active People. *International Journal of Sports Medicine*, *36*(05), 426–430.

de Lourdes Nahhas Rodacki, C., Rodacki, A. L. F., Coelho, I., Pequito, D., Krause, M., Bonatto, S., … Fernandes, L. C. (2015). Influence of fish oil supplementation and strength training on some functional aspects of immune cells in healthy elderly women. *The British Journal of Nutrition*, *114*(1), 43–52.

DiLorenzo, F. M., Drager, C. J., & Rankin, J. W. (2014). Docosahexaenoic acid affects markers of inflammation and muscle damage after eccentric exercise. *Journal of Strength and Conditioning Research*, *28*(10), 2768–2774.

Djoussé, L., Bartz, T. M., Ix, J. H., Kochar, J., Kizer, J. R., Gottdiener, J. S., … Zieman, S. J. (2013). Fatty acid-binding protein 4 and incident heart failure: the Cardiovascular Health Study. *European Journal of Heart Failure*, *15*(4), 394–399.

Eston, R. G., Finney, S., Baker, S., & Baltzopoulos, V. (1996). Muscle tenderness and peak torque changes after downhill running following a prior bout of isokinetic eccentric exercise. *Journal of Sports Sciences*, *14*(4), 291–299.

Eyres, L., Eyres, M. F., Chisholm, A., & Brown, R. C. (2016). Coconut oil consumption and cardiovascular risk factors in humans. *Nutrition Reviews*, *74*(4), 267–280.

Fontani, G., Corradeschi, F., Felici, A., Alfatti, F., Migliorini, S., & Lodi, L. (2005). Cognitive and physiological effects of Omega-3 polyunsaturated fatty acid supplementation in healthy subjects. *European Journal of Clinical Investigation*, *35*(11), 691–699.

Glover, E. I., Phillips, S. M., Oates, B. R., Tang, J. E., Tarnopolsky, M. A., Selby, A., Smith, K., Rennie, M. J. (2008). Immobilization induces anabolic resistance in human

myofibrillar protein synthesis with low and high dose amino acid infusion. *The Journal of Physiology*, *586*(24), 6049-6061.

Gravina, L., Brown, F. F., Alexander, L., Dick, J., Bell, G., Witard, O. C., & Galloway, S. D. R. (2017). 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. *International Journal of Sport Nutrition and Exercise Metabolism*, *27*(4), 305–313.

Gray, P., Chappell, A., Jenkinson, A. M., Thies, F., & Gray, S. R. (2014). Fish Oil Supplementation Reduces Markers of Oxidative Stress but Not Muscle Soreness after Eccentric Exercise. *International Journal of Sport Nutrition and Exercise Metabolism*, *24*(2), 206–214.

Hancock, C. R., Han, D.-H., Chen, M., Terada, S., Yasuda, T., Wright, D. C., & Holloszy, J. O. (2008). High-fat diets cause insulin resistance despite an increase in muscle mitochondria. *Proceedings of the National Academy of Sciences of the United States of America*, *105*(22), 7815–7820.

Hingley, L., Macartney, M. J., Brown, M. A., McLennan, P. L., & Peoples, G. E. (2017). DHA-rich Fish Oil Increases the Omega-3 Index and Lowers the Oxygen Cost of Physiologically Stressful Cycling in Trained Individuals. *International Journal of Sport Nutrition and Exercise Metabolism*, *27*(4), 335–343.

Hooper, L., Thompson, R. L., Harrison, R. A., Summerbell, C. D., Ness, A. R., Moore, H. J., … Smith, G. D. (2006). Risks and benefits of omega 3 fats for mortality, cardiovascular disease, and cancer: systematic review. *BMJ*, *332*(7544), 752–760.

Hoppeler, H., Lüthi, P., Claassen, H., Weibel, E. R., & Howald, H. (1973). The ultrastructure of the normal human skeletal muscle. A morphometric analysis on untrained men,

women and well-trained orienteers. *Pflugers Archiv : European Journal of Physiology*, *344*(3), 217–232.

Huang, S., & Czech, M. P. (2007). The GLUT4 Glucose Transporter. *Cell Metabolism*, *5*(4), 237–252.

Hulbert, A. J., Turner, N., Storlien, L. H., & Else, P. L. (2005). Dietary fats and membrane function: implications for metabolism and disease. *Biological Reviews of the Cambridge Philosophical Society*, *80*(1), 155–169.

Jakeman, J. R., Lambrick, D. M., Wooley, B., Babraj, J. A., & Faulkner, J. A. (2017). Effect of an acute dose of omega-3 fish oil following exercise-induced muscle damage. *European Journal of Applied Physiology*, *117*(3), 575–582.

Janakiram, N. B., Mohammed, A., & Rao, C. V. (2011). Role of lipoxins, resolvins, and other bioactive lipids in colon and pancreatic cancer. *Cancer and Metastasis Reviews*, *30*(3–4), 507–523.

Jouris, K. B., McDaniel, J. L., & Weiss, E. P. (2011). The Effect of Omega-3 Fatty Acid Supplementation on the Inflammatory Response to eccentric strength exercise. *Journal of Sports Science & Medicine*, *10*(3), 432–438.

Kamolrat, T., & Gray, S. R. (2013). The effect of eicosapentaenoic and docosahexaenoic acid on protein synthesis and breakdown in murine C2C12 myotubes. *Biochemical and Biophysical Research Communications*, *432*(4), 593–598.

Kawabata, F., Neya, M., Hamazaki, K., Watanabe, Y., Kobayashi, S., & Tsuji, T. (2014). Supplementation with eicosapentaenoic acid-rich fish oil improves exercise economy and reduces perceived exertion during submaximal steady-state exercise in normal healthy untrained men. *Bioscience, Biotechnology, and Biochemistry*, *78*(12), 2081–2088.

Kim, H.-Y., Huang, B. X., & Spector, A. A. (2014). Phosphatidylserine in the brain: Metabolism and function. *Progress in Lipid Research*, *56*, 1–18.

Laiglesia, L. M., Lorente-Cebrián, S., Prieto-Hontoria, P. L., Fernández-Galilea, M., Ribeiro, S. M. R., Sáinz, N., … Moreno-Aliaga, M. J. (2016). Eicosapentaenoic acid promotes mitochondrial biogenesis and beige-like features in subcutaneous adipocytes from overweight subjects. *The Journal of Nutritional Biochemistry*, *37*, 76–82.

Lanza, I. R., Blachnio-Zabielska, A., Johnson, M. L., Schimke, J. M., Jakaitis, D. R., Lebrasseur, N. K., … Zabielski, P. (2013). Influence of fish oil on skeletal muscle mitochondrial energetics and lipid metabolites during high-fat diet. *American Journal of Physiology-Endocrinology and Metabolism*, *304*(12), E1391–E1403.

Lee, T. H., Hoover, R. L., Williams, J. D., Sperling, R. I., Ravalese, J., Spur, B. W., … Austen, K. F. (1985). Effect of Dietary Enrichment with Eicosapentaenoic and Docosahexaenoic Acids on in Vitro Neutrophil and Monocyte Leukotriene Generation and Neutrophil Function. *New England Journal of Medicine*, *312*(19), 1217–1224.

Lembke, P., Capodice, J., Hebert, K., & Swenson, T. (2014). Influence of omega-3 (n3) index on performance and wellbeing in young adults after heavy eccentric exercise. *Journal of Sports Science & Medicine*, *13*(1), 151–156.

Li, H., Ruan, X. Z., Powis, S. H., Fernando, R., Mon, W. Y., Wheeler, D. C., … Varghese, Z. (2005). EPA and DHA reduce LPS-induced inflammation responses in HK-2 cells: Evidence for a PPAR-γ–dependent mechanism. *Kidney International*, *67*(3), 867–874.

Lim, K., Han, C., Dai, Y., Shen, M., & Wu, T. (2009). Omega-3 polyunsaturated fatty acids inhibit hepatocellular carcinoma cell growth through blocking -catenin and cyclooxygenase-2. *Molecular Cancer Therapeutics*, *8*(11), 3046–3055.

Logan, S. L., & Spriet, L. L. (2015). Omega-3 Fatty Acid Supplementation for 12 Weeks Increases Resting and Exercise Metabolic Rate in Healthy Community-Dwelling Older Females. *PLOS ONE*, *10*(12).

Longland, T. M., Oikawa, S. Y., Mitchell, C. J., Devries, M. C., & Phillips, S. M. (2016). Higher compared with lower dietary protein during an energy deficit combined with intense exercise promotes greater lean mass gain and fat mass loss: a randomized trial. *The American Journal of Clinical Nutrition*, *103*(3), 738–746.

Lorenz, R., Spengler, U., Fischer, S., Duhm, J., & Weber, P. C. (1983). Platelet function, thromboxane formation and blood pressure control during supplementation of the Western diet with cod liver oil. *Circulation*, *67*(3), 504–511.

Macartney, M. J., Hingley, L., Brown, M. A., Peoples, G. E., & McLennan, P. L. (2014). Intrinsic heart rate recovery after dynamic exercise is improved with an increased omega-3 index in healthy males. *British Journal of Nutrition*, *112*(12), 1984–1992.

Marik, P. E., & Varon, J. (2009). Omega-3 Dietary Supplements and the Risk of Cardiovascular Events: A Systematic Review. *Clinical Cardiology*, *32*(7), 365–372.

Maughan, R. J., Depiesse, F., Geyer, H., & International Association of Athletics Federations. (2007). The use of dietary supplements by athletes. *Journal of Sports Sciences*, *25*(sup1), S103–S113.

McEwen, B., Morel-Kopp, M.-C., Chen, W., Tofler, G., & Ward, C. (2013). Effects of Omega-3 Polyunsaturated Fatty Acids on Platelet Function in Healthy Subjects and Subjects with Cardiovascular Disease. *Seminars in Thrombosis and Hemostasis*, *39*(01), 25–32.

McGlory, C., Galloway, S. D. R., Hamilton, D. L., McClintock, C., Breen, L., Dick, J. R., … Tipton, K. D. (2014). Temporal changes in human skeletal muscle and blood lipid

composition with fish oil supplementation. *Prostaglandins, Leukotrienes and Essential Fatty Acids (PLEFA)*, *90*(6), 199–206.

McGlory, C., Wardle, S. L., Macnaughton, L. S., Witard, O. C., Scott, F., Dick, J., … Tipton, K. D. (2016). Fish oil supplementation suppresses resistance exercise and feeding-induced increases in anabolic signaling without affecting myofibrillar protein synthesis in young men. *Physiological Reports*, *4*(6), e12715.

McKinley-Barnard, S. K., Andre, T. L., Gann, J. J., Hwang, P. S., & Willoughby, D. S. (2018). Effectiveness of Fish Oil Supplementation in Attenuating Exercise-Induced Muscle Damage in Women During Midfollicular and Midluteal Menstrual Phases. *Journal of Strength and Conditioning Research*, *32*(6), 1601–1612.

Mettler, S., Mitchell, N., & Tipton, K. D. (2010). Increased Protein Intake Reduces Lean Body Mass Loss during Weight Loss in Athletes. *Medicine & Science in Sports & Exercise*, *42*(2), 326–337.

Mozaffarian, D., Gottdiener, J. S., & Siscovick, D. S. (2006). Intake of tuna or other broiled or baked fish versus fried fish and cardiac structure, function, and hemodynamics. *The American Journal of Cardiology, 97*(2), 216-222.

Murphy, M. G. (1990). Dietary fatty acids and membrane protein function. *The Journal of Nutritional Biochemistry*, *1*(2), 68–79.

Murphy, R. A., Mourtzakis, M., Chu, Q. S. C., Baracos, V. E., Reiman, T., & Mazurak, V. C. (2011). Nutritional intervention with fish oil provides a benefit over standard of care for weight and skeletal muscle mass in patients with nonsmall cell lung cancer receiving chemotherapy. *Cancer*, *117*(8), 1775–1782.

Nédélec, M., McCall, A., Carling, C., Legall, F., Berthoin, S., & Dupont, G. (2012).

    Recovery in Soccer. *Sports Medicine*, *42*(12), 997–1015.

Ninio, D. M., Hill, A. M., Howe, P. R., Buckley, J. D., & Saint, D. A. (2008).

    Docosahexaenoic acid-rich fish oil improves heart rate variability and heart rate

    responses to exercise in overweight adults. *British Journal of Nutrition*, *100*(05),

    1097.

O'Keefe, J. H. Jr., Abuissa, H., Sastre, A., Steinhaus, D. M., and Harris, W. S. (2006). Effects

    of omega3 fatty acids on resting heart rate, heart rate recovery after exercise, and

    heart rate variability in men with healed myocardial infarctions and depressed

    ejection fractions. *The American Journal of Cardiology, 97*(8), 1127–1130.

Oliver, J. M., Jones, M. T., Kirk, K. M., Gable, D. A., Repshas, J. T., Johnson, T. A., …

    Zetterberg, H. (2016). Effect of Docosahexaenoic Acid on a Biomarker of Head

    Trauma in American Football. *Medicine & Science in Sports & Exercise*, *48*(6),

    974–982.

Oostenbrug, G. S., Mensink, R. P., Hardeman, M. R., De Vries, T., Brouns, F., & Hornstra,

    G. (1997). Exercise performance, red blood cell deformability, and lipid

    peroxidation: effects of fish oil and vitamin E. *Journal of Applied Physiology*, *83*(3),

    746–752.

Papanikolaou, Y., Brooks, J., Reider, C., & Fulgoni, V. L. (2014). U.S. adults are not meeting

    recommended levels for fish and omega-3 fatty acid intake: results of an analysis

    using observational data from NHANES 2003-2008. *Nutrition Journal*, *13*(1), 31.

Pasiakos, S. M., Vislocky, L. M., Carbone, J. W., Altieri, N., Konopelski, K., Freake, H. C.,

    … Rodriguez, N. R. (2010). Acute Energy Deprivation Affects Skeletal Muscle

    Protein Synthesis and Associated Intracellular Signaling Proteins in Physically

    Active Adults. *The Journal of Nutrition*, *140*(4), 745–751.

Peoples, G. E., McLennan, P. L., Howe, P. R. C., & Groeller, H. (2008). Fish Oil Reduces Heart Rate and Oxygen Consumption During Exercise. *Journal of Cardiovascular Pharmacology*, *52*(6), 540–547.

Pereira Panza, V. S., Diefenthaeler, F., & da Silva, E. L. (2015). Benefits of dietary phytochemical supplementation on eccentric exercise-induced muscle damage: Is including antioxidants enough? *Nutrition*, *31*(9), 1072–1082.

Peters, E. M., & Bateman, E. D. (1983). Ultramarathon running and upper respiratory tract infections. An epidemiological survey. *South African Medical Journal = Suid-Afrikaanse Tydskrif Vir Geneeskunde*, *64*(15), 582–584.

Philpott, J. D., Donnelly, C., Walshe, I. H., MacKinley, E. E., Dick, J., Galloway, S. D. R., … Witard, O. C. (2018). Adding Fish Oil to Whey Protein, Leucine, and Carbohydrate Over a Six-Week Supplementation Period Attenuates Muscle Soreness Following Eccentric Exercise in Competitive Soccer Players. *International Journal of Sport Nutrition and Exercise Metabolism*, *28*(1), 26–36.

Rodacki, C. L., Rodacki, A. L., Pereira, G., Naliwaiko, K., Coelho, I., Pequito, D., & Fernandes, L. C. (2012). Fish-oil supplementation enhances the effects of strength training in elderly women. *The American Journal of Clinical Nutrition*, *95*(2), 428–436.

Rogerson, D. (2017). Vegan diets: practical advice for athletes and exercisers. *Journal of the International Society of Sports Nutrition*, *14*(1), 36.

Rontoyanni, V. G., Hall, W. L., Pombo-Rodrigues, S., Appleton, A., Chung, R., & Sanders, T. A. B. (2012). A comparison of the changes in cardiac output and systemic vascular resistance during exercise following high-fat meals containing DHA or EPA. *British Journal of Nutrition*, *108*(03), 492–499.

Shaw, G., Slater, G., & Burke, L. M. (2016). Changes in the Supplementation Practices of Elite Australian Swimmers Over 11 Years. *International Journal of Sport Nutrition and Exercise Metabolism*, *26*(6), 565–571.

Smith, G. I., Atherton, P., Reeds, D. N., Mohammed, B. S., Rankin, D., Rennie, M. J., & Mittendorfer, B. (2011a). Dietary omega-3 fatty acid supplementation increases the rate of muscle protein synthesis in older adults: a randomized controlled trial. *The American Journal of Clinical Nutrition*, *93*(2), 402–412.

Smith, G. I., Julliand, S., Reeds, D. N., Sinacore, D. R., Klein, S., & Mittendorfer, B. (2015). Fish oil–derived n−3 PUFA therapy increases muscle mass and function in healthy older adults1. *The American Journal of Clinical Nutrition*, *102*(1), 115–122.

Smith, G. I., Atherton, P., Reeds, D. N., Mohammed, B. S., Rankin, D., Rennie, M. J., & Mittendorfer, B. (2011b). Omega-3 polyunsaturated fatty acids augment the muscle protein anabolic response to hyperinsulinaemia–hyperaminoacidaemia in healthy young and middle-aged men and women. *Clinical Science*, *121*(6), 267–278.

Sprague, M., Dick, J. R., & Tocher, D. R. (2016). Impact of sustainable feeds on omega-3 long-chain fatty acid levels in farmed Atlantic salmon, 2006–2015. *Scientific Reports*, *6*(1), 21892.

Tartibian, B., Maleki, B. H., & Abbasi, A. (2011). Omega-3 Fatty Acids Supplementation Attenuates Inflammatory Markers After Eccentric Exercise in Untrained Men. *Clinical Journal of Sport Medicine*, *21*(2), 131–137.

Tsuchiya, Y., Yanagimoto, K., Nakazato, K., Hayamizu, K., & Ochi, E. (2016). Eicosapentaenoic and docosahexaenoic acids-rich fish oil supplementation attenuates strength loss and limited joint range of motion after eccentric contractions: a randomized, double-blind, placebo-controlled, parallel-group trial. *European Journal of Applied Physiology*, *116*(6), 1179–1188.

Turner, N., Bruce, C. R., Beale, S. M., Hoehn, K. L., So, T., Rolph, M. S., & Cooney, G. J. (2007). Excess Lipid Availability Increases Mitochondrial Fatty Acid Oxidative Capacity in Muscle. *Diabetes*, *56*(8), 2085–2092.

Wall, B. T., & van Loon, L. J. C. (2013). Nutritional strategies to attenuate muscle disuse atrophy. *Nutrition Reviews, 71*(4), 195-208.

Wang, T., Van, K. C., Gavitt, B. J., Grayson, J. K., Lu, Y.-C., Lyeth, B. G., & Pichakron, K. O. (2013). Effect of fish oil supplementation in a rat model of multiple mild traumatic brain injuries. *Restorative Neurology and Neuroscience*, *31*(5), 647–59.

Watt, M. J., Heigenhauser, G. J. F., Dyck, D. J., & Spriet, L. L. (2002). Intramuscular triacylglycerol, glycogen and acetyl group metabolism during 4 h of moderate exercise in man. *The Journal of Physiology*, *541*(Pt 3), 969–78.

Weinheimer, E. M., Sands, L. P., & Campbell, W. W. (2010). A systematic review of the separate and combined effects of energy restriction and exercise on fat-free mass in middle-aged and older adults: implications for sarcopenic obesity. *Nutrition Reviews*, *68*(7), 375–388.

Witard, O. C., Jackman, S. R., Breen, L., Smith, K., Selby, A., & Tipton, K. D. (2014). Myofibrillar muscle protein synthesis rates subsequent to a meal in response to increasing doses of whey protein at rest and after resistance exercise. *The American Journal of Clinical Nutrition*, *99*(1), 86–95.

World Health Organization. (2003) Expert Consultation on Diet, Nutrition and the Prevention of Chronic Diseases. *Expert Consultation on Diet, Nutrition and the Prevention of Chronic Diseases*. Geneva, Switzerland.

You, J.-S., Park, M.-N., & Lee, Y.-S. (2010b). Dietary fish oil inhibits the early stage of

    recovery of atrophied soleus muscle in rats via Akt–p70s6k signaling and PGF2α.

    The Journal of Nutritional Biochemistry, *21*(10), 929–934.

You, J.-S., Park, M.-N., Song, W., & Lee, Y.-S. (2010a). Dietary fish oil alleviates soleus

    atrophy during immobilization in association with Akt signaling to p70s6k and E3

    ubiquitin ligases in rats. *Applied Physiology, Nutrition, and Metabolism*, *35*(3), 310–

    318.

Żebrowska, A., Mizia-Stec, K., Mizia, M., Gąsior, Z., & Poprzęcki, S. (2015). Omega-3 fatty

    acids supplementation improves endothelial function and maximal oxygen uptake in

    endurance-trained athletes. *European Journal of Sport Science*, *15*(4), 305–14.

Zucker, M. L., Bilyeu, D. S., Helmkamp, G. M., Harris, W. S., & Dujovne, C. A. (1988).

    Effects of dietary fish oil on platelet function and plasma lipids in

    hyperlipoproteinemic and normal subjects. *Atherosclerosis*, *73*(1), 13–22.