Metabolic Health Benefits of Long-Chain Omega-3 Polyunsaturated Fatty Acids

Peter Howe, PhD†; Jon Buckley, PhD**

ABSTRACT Restricting energy intake and increasing physical activity are advocated for reducing obesity, but many individuals have difficulty complying with these recommendations. Consumption of long-chain omega-3 polyunsaturated fatty acids (n-3 LCPUFA) offers multiple mechanisms to counteract obesity, including appetite suppression; circulatory improvements, which promote nutrient delivery to skeletal muscle and changes in gene expression, which shift metabolism toward increased fat oxidation; increased energy expenditure; and reduced fat deposition. n-3 LCPUFA may also alter gene expression in skeletal muscle to suppress catabolic pathways and upregulate anabolic pathways, resulting in greater lean tissue mass, metabolic rate, and maintenance of physical function. n-3 LCPUFA supplementation has been shown to counteract obesity in rodents, but evidence in humans is limited. Epidemiological associations between n-3 LCPUFA intakes and obesity are inconclusive. Several studies, on the other hand, indicate inverse relationships between biomarkers of n-3 LCPUFA status and obesity, although causality is uncertain. There have been few human intervention trials of omega-3 supplementation for obesity; some have indicated potential benefits, especially when combined with energy-restricted diets or exercise. More trials are needed to confirm these effects and identify mechanisms of action.

INTRODUCTION

The growing prevalence of obesity throughout the developed world is bringing an increased risk of metabolic and cardiovascular disease. Current recommendations for reducing obesity are based on dietary energy restriction and increased physical activity, but many individuals have difficulty complying with these recommendations and additional strategies to assist body fat reduction are required. Increased intake of the long-chain omega-3 polyunsaturated fatty acids (n-3 LCPUFA) eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) may offer a more acceptable alternative.

EPIDEMIOLOGICAL STUDIES

A number of large prospective studies examining effects of n-3 LCPUFA supplementation on cardiovascular health have included data on body mass index (BMI). Baseline data in 43,671 men from the Health Professional Follow-Up Study[1] indicated that men with high fish consumption were less likely to be overweight than those with low fish consumption, and the proportion of overweight volunteers was inversely related to n-3 LCPUFA intake. Conversely, the Nurses' Health Study,[2] which examined the relationship between fish and n-3 LCPUFA intake on stroke risk in 79,839 women, reported that higher intakes of fish and n-3 LCPUFA were associated with a higher prevalence of obesity (based on BMI). Although the higher prevalence of obesity associated with fish intake could be accounted for by a higher energy intake, this was not the case for n-3 LCPUFA intake. Thus, these two large studies provide conflicting evidence on the association between n-3 LCPUFA intake and obesity. However, both estimated dietary intakes from food frequency questionnaires, which are only semiquantitative and may be limited in their ability to accurately assess intakes of different types of fat.

An alternative approach is to assess a surrogate biomarker of n-3 LCPUFA intake. Micaleff et al[3] measured concentrations of n-3 LCPUFA in plasma phospholipids of 124 adults and tested for relationships with markers of obesity. It was found that n-3 LCPUFA as a percentage of total fatty acids in plasma phospholipids was inversely related to BMI, waist circumference, and hip circumference. This is consistent with the observation of significantly lower plasma phospholipid DHA level in obese versus normal weight adolescents,[4] and a small-scale study showing that adipose tissue in adolescent girls is inversely related to n-3 LCPUFA in plasma phospholipids.[5]

Fatty acid levels in plasma phospholipids reflect uptake and retention of dietary fatty acids over a period of weeks (as opposed to plasma fatty acids which reflect intake in recent days); hence, they are regarded as a surrogate marker of habitual dietary intake. However, the most consistent indicator of the uptake and incorporation of fatty acids in tissues is the relative content of erythrocytes, where DHA, for example, is retained in the plasma membrane for the 4-month life of the cell.[6] We examined erythrocyte fatty acid data obtained at baseline from 476 adult volunteers participating in various nutritional intervention trials in our own laboratory and found that erythrocyte n-3 LCPUFA levels were inversely associated with BMI, waist circumference, and percentage body fat assessed by dual-energy X-ray absorptiometry (DEXA).[7] Consistent with the observations in adolescents, these associations

*Alliance for Research in Exercise, Nutrition and Activity (ARENA), Sansom Institute for Health Research, University of South Australia, Adelaide, South Australia 5000, Australia.

†Clinical Nutrition Research Centre, University of Newcastle, Callaghan, New South Wales 2308, Australia.

doi: 10.7205/MILMED-D-14-00154

were primarily attributable to erythrocyte DHA content and were stronger for women than men, reflecting a possible gender difference in the influence of omega-3 status on adiposity.

Thus, evidence from cross-sectional studies (albeit relatively small sample sizes) indicates that higher erythrocyte levels (suggestive of higher habitual intakes) of n-3 LCPUFA, particularly DHA in females, are associated with less adiposity.

INTERVENTION TRIALS

Numerous intervention trials have examined the influence of n-3 LCPUFA on adiposity in experimental animals, with fairly consistent results. Increased intakes of n-3 LCPUFA can reduce body fat accumulation in a dose-dependent manner in rodents exposed to a high-fat obesogenic diet[8–13] and reduce body fat in rodents that are already obese.[14] Some[11,13] but not all[9,10,12,14] of these studies demonstrated that the reduction in body fat was associated with a reduction in food intake, suggesting a possible effect of n-3 LCPUFA consumption on appetite. Although n-3 LCPUFA appear to improve body composition in rodents, the impact of an increased n-3 LCPUFA intake on body composition in humans is less certain as few data are available and that which is available is conflicting, possibly due to poor study design and/or lack of appropriate controls. Human intervention trials have administered n-3 LCPUFA as fish oil alone or in combination with energy-restricted diets and/or exercise interventions and are categorized accordingly below.

Effects of n-3 LCPUFA Intake Alone

The earliest reported clinical investigation of effects of n-3 LCPUFA on adiposity by Couet et al[15] had volunteers consume a control diet for 3 weeks and then, after a 10- to 12-week washout, consume the same diet with 6 g/d of visible fat replaced with fish oil. Resting energy expenditure and resting fat oxidation were increased and body fat (assessed by DEXA) was reduced on the fish oil diet compared with control. However, this outcome could have been due to an order effect, which may have confounded the outcome; there was no parallel control group.

Groh-Wargo et al[16] fed preterm infants a formula supplemented with arachidonic acid and DHA to 12 months of gestation-corrected age and reported less body fat accumulation compared with controls. Although this finding is consistent with n-3 LCPUFA supplementation reducing fat accumulation in growing rats fed an obesogenic diet, the study design did not allow for determination of whether the effect was mediated by DHA or arachidonic acid as both were provided together.

Kabir et al[17] randomized overweight/obese female postmenopausal volunteers with type 2 diabetes to consume 3 g/d of EPA-rich fish oil or 3 g/d of paraffin oil for 2 months. Neither treatment reduced body weight, but n-3 LCPUFA reduced total body fat mass, primarily due to the loss of trunk fat, which was associated with a reduction of adipocyte diameter in subcutaneous periumbilical fat. However, an earlier study by Todoric et al[18] in genetically diabetic adult mice found that consumption of n-3 LCPUFA increased body fat. The reason for this difference is not clear, although body composition is likely to be regulated differently in mice that are genetically predisposed to diabetes compared with humans who develop type 2 diabetes as a result of lifestyle habits.

Chrochemore et al[19] recently claimed a reduction in body weight in a dose–response study of fish oil in patients with type 2 diabetes. However, their trial was of short duration (4 weeks) with low doses of fish oil (<1 g/d) and there was no evidence of a statistically significant treatment effect. Rafraf et al[20] conducted a clinical trial in women with polycystic ovary syndrome over an 8-week period with 1.2 g/d of fish oil but also failed to detect any improvement in measures of adiposity.

Thus, a few relatively small studies in humans have examined effects of n-3 LCPUFA supplementation alone on adiposity and the variability of outcomes can be attributed largely to limitations in study design. The limited evidence available suggests that an increased intake of n-3 LCPUFA may reduce body fat. However, additional studies are required in both obesity and type 2 diabetes to define the effects of n-3 LCPUFA as a sole intervention on body composition.

Effects of n-3 LCPUFA Intake Combined With Exercise

Regular exercise and dietary energy restriction are cornerstones in the management of obesity, and a number of studies have examined whether combining an increased n-3 LCPUFA intake with these lifestyle changes can enhance weight loss.

Early studies examined whether an increased intake of n-3 LCPUFA in combination with exercise could provide greater improvements in body composition than exercise alone. Warner et al[21] randomized 34 obese adults to consume 50 mL/d of corn oil (control), 50 mL/d of fish oil, or 50 mL/d of fish oil combined with moderate intensity aerobic exercise (walking or jogging) three times per week. Volunteers who exercised and consumed fish oil experienced a 3% reduction in body fat (assessed by DEXA), whereas body fat was unchanged in the other treatment groups. However, the study design did not make it possible to determine whether the reduction in body fat in the combined intervention group was due to the exercise alone or to some interaction between the fish oil and exercise; the lack of any change in body fat in the fish oil only treatment group suggests that the former was most likely. This limitation in study design was addressed in a subsequent study,[22] where 32 healthy sedentary young males were randomized to control fish oil, exercise, or fish oil and exercise treatments for 10 weeks. However, body fat did not change in any treatment group, probably because the volunteers were lean at study commencement and this may have limited their capacity to reduce body fat.

We examined effects of n-3 LCPUFA and exercise independently and in combination on body composition by randomizing 65 overweight/obese adults to consume DHA-rich tuna oil or sunflower oil for 12 weeks in a 2 × 2 factorial parallel design trial.[23] Subjects in each treatment group were further randomized to maintain their usual physical activity or undertake moderate intensity aerobic exercise (jogging/walking) three times per week. Both exercise and n-3 LCPUFA supplementation independently reduced body fat and their effects were additive, such that volunteers combining fish oil supplementation with regular exercise reduced body fat by ~1.5 kg.

Although dietary energy restriction is an effective means for reducing body fat, it typically results also in the loss of lean tissue including skeletal muscle which, apart from potentially impacting negatively on physical function, can also reduce metabolic rate and oppose further body fat reduction. The pathway predominantly responsible for skeletal muscle catabolism during energy restriction is the ubiquitin–proteasome pathway.[24] EPA has been shown to suppress the activity of this pathway during severe dietary energy restriction,[25] and n-3 LCPUFA have also been shown in animals to increase activation of the anabolic Akt–mTOR–S6K1 pathway in skeletal muscle.[26] The ability of n-3 LCPUFA to reduce catabolic activity and increase anabolic activity in skeletal muscle suggests that these fatty acids may protect against the loss of skeletal muscle during energy-restricted weight loss and thus maintain metabolic rate, promoting further weight loss and assisting in the maintenance of physical function. Indeed, data from our laboratory indicate that erythrocyte n-3 LCPUFA levels are positively associated with percentage lean body mass (assessed by DEXA),[7] suggesting that an increased intake of n-3 LCPUFA might stimulate an increase in lean tissue mass.

Effects of n-3 LCPUFA Intake Combined With Energy Restriction

Although no studies to date have directly examined the effect of increased n-3 LCPUFA intake on the maintenance of lean tissue mass during energy-restricted weight loss, a number have studied the effects on body weight and body fat of consuming these fatty acids in combination with an energy-restricted diet. Fontani et al[27] examined the effects of n-3 LCPUFA supplementation with fish oil versus olive oil against a background of two different energy-controlled diets on weight loss in noncompetitive athletes but found no additional effect of supplementing with n-3 LCPUFA on weight loss or reductions in body fat. However, this study assessed changes in body fat using skinfolds, did not control or monitor physical activity levels during the intervention, and used a crossover design with no washout. It has been shown that the increased incorporation of n-3 LCPUFA into erythrocyte membranes (a marker of longer-term bioavailability) is sustained for up to 18 weeks.[6] Thus, any metabolic effects resulting from tissue incorporation of n-3 LCPUFA in volunteers supplemented with n-3 LCPUFA during the first 5-week dosing period may have persisted during the second 5-week period when an olive oil control was consumed, potentially confounding the study outcomes.

Krebs et al[28] added n-3 LCPUFA to a low-fat high-carbohydrate weight-loss diet in overweight women for 12 weeks (followed by 12 weeks of a weight maintenance diet combined with n-3 LCPUFA) and found no additional reduction in body weight or body fat. However, as in the study by Fontani et al,[27] volunteers undertook exercise in conjunction with the dietary energy restriction but the amount of exercise undertaken was not monitored or controlled, thus potentially confounding the outcome. Interestingly, although the study by Krebs et al did not demonstrate any benefit of n-3 LCPUFA for improving body composition, it was found that n-3 LCPUFA supplementation attenuated the decrease in plasma concentrations of the appetite-suppressing hormone leptin that occur with weight loss. This latter finding is consistent with the study by Pérez-Matute et al[13] in rats, which showed that n-3 LCPUFA intake was associated with a reduced accumulation of body fat when exposed to a high-fat obesogenic diet and that this attenuation of body fat accumulation was associated with increased plasma leptin concentrations and suppressed appetite. Thus, n-3 LCPUFA might assist in maintaining circulating leptin concentrations during energy-restricted weight loss, which may suppress appetite and assist with compliance to an energy-restricted diet.

Thorsdottir et al[29] randomized overweight volunteers to one of four isocaloric energy-restricted diets for 8 weeks: a control diet with no seafood, a diet containing lean fish, a diet containing fatty fish rich in n-3 LCPUFA, or a diet supplemented with 6 fish oil capsules per day. They found that weight loss and reductions in waist circumference were greater in the groups receiving fish or fish oil compared with controls. Again, however, physical activity levels were not monitored or controlled, making it difficult to interpret whether the greater improvements in body composition were due to the diets or to potentially confounding differences in physical activity. Interestingly, a subanalysis of data from the final 2 weeks of this study showed greater satiety following consumption of meals rich in n-3 LCPUFA (i.e., fatty fish and fish oil capsules) compared with the control or lean fish diets,[30] suggesting an appetite-suppressing effect of n-3LCPUFA.

Several studies which combined n-3 LCPUFA supplementation with dietary energy restriction also controlled physical activity levels during the intervention. One such study was conducted in a hospital metabolic ward.[31] This inpatient study randomized severely obese women to consume either n-3 LCPUFA or placebo in conjunction with a very-low-energy diet (VLED) and regular exercise for 3 weeks. The patients who consumed the n-3 LCPUFA experienced greater reductions in BMI and hip circumference and a trend toward greater weight loss compared with controls, but effects on body fat or

lean mass were not assessed. Nevertheless, the findings of this study indicate that, when physical activity is controlled, an increased n-3 LCPUFA intake during dietary energy restriction results in greater improvements in markers of obesity.

DeFina et al[32] attempted to see if n-3 LCPUFA supplementation could augment the effects of a diet and exercise program for weight loss. They saw no significant differences between fish oil (1 g EPA, 0.2 g DHA daily) and placebo in the reduction of BMI, waist circumference, or % body fat over a 6-month period. However, variations in background energy balance are a potential confounder when ascribing outcomes to the supplementation; although not significant, there was a greater reduction of energy intake in the placebo group. Physical activity levels appeared similar.

Most recently, Munro and Garg[33–35] have reported a series of three studies, in which the evaluated effects on measures of adiposity of administering DHA-rich tuna oil either before or during consumption of either an energy reduced or a VLED. When fish oil administration preceded commencement of the VLED, weight loss was greater than in the control group.[34] However, the other studies also tended to show greater weight loss with fish oil treatment; both were small and may have been underpowered to individually reach statistical significance.[33,35]

Thus, a number of studies have assessed the effects of n-3 LCPUFA supplementation on body weight and/or body composition during dietary energy restriction with conflicting results. Only one controlled for the potentially confounding effects of physical activity[31] and this study showed an additional benefit of n-3 LCPUFA. None of these studies specifically evaluated whether n-3 LCPUFA supplementation provided any protection against the loss of lean tissue. There is a need for longer term well-controlled studies to evaluate whether n-3 LCPUFA can provide greater reductions in body fat and protect against the loss of lean tissue when consumed with an energy-restricted diet and/or exercise.

POSSIBLE MECHANISMS OF ACTION

Adipokines, Appetite Suppression, and Thermogenesis

The mechanisms by which n-3 LCPUFA may reduce body fat are not well understood. Increasing evidence from human[30] and animal[11,13] studies suggest that these fatty acids may suppress appetite and/or increase thermogenesis, possibly by increasing plasma concentrations of adiponectin and leptin,[13,28] the latter particularly in obese individuals.[36] However, not all studies which have reported reductions in body fat have shown any effect on food intake.

Gene Expression

There is considerable evidence from animal studies that the reductions in body fat that occur with increased n-3 LCPUFA intake are in-part mediated by altering the expression of genes regulating fat metabolism in a number of tissues. Feeding n-3 LCPUFA has been shown to upregulate the expression of genes and proteins involved in fatty acid oxidation in liver, intestine, cardiac and skeletal muscles, and adipose tissue[10,37–39] and downregulate the expression of genes involved in lipogenesis in adipose tissue,[38] resulting in a shift in metabolic profile toward one which favors fat oxidation and reduced fat storage. In a recent in vitro study, Barber et al[40] found that DHA specifically increased lipolysis and reduced the gene expression of three proteins related to lipid droplet formation in adipocytes.

There is also some limited evidence which suggests that n-3 LCPUFA may promote body fat loss via alterations in the activity of anticatabolic and/or anabolic pathways in skeletal muscle, which promote the maintenance of lean tissue mass and metabolic rate. EPA suppresses activation of the ubiquitin–proteasome pathway,[25] which is the key pathway responsible for muscle proteolysis during energy restriction[24] by impairing activation of the transcription factor NF-κβ, an important regulator of the proteasomal pathway.[25,41] Animal studies have also shown that n-3 LCPUFA increase whole-body protein synthesis via the activation of key regulatory kinases involved in muscle protein synthesis, specifically mTOR and S6K.[26] These anticatabolic and anabolic effects of n-3 LCPUFA may attenuate muscle loss and promote the maintenance of muscle mass (and metabolic rate) during periods of dietary energy restriction or potentially increase muscle mass during energy balance. However, two recent studies[42,43] in athletes which assessed the potential benefits of supplementation with n-3 LCPUFA for improving physical performance during endurance exercise activities reported no additional benefit of n-3 LCPUFA supplementation compared with control, suggesting no beneficial effect on skeletal muscle anabolism. Additional studies of the potential for n-3 LCPUF to maintain lean tissue mass during energy restriction appear warranted.

Circulatory Effects

Arterial vasodilator function, as assessed by flow-mediated dilatation of the brachial artery[44–46] and skeletal muscle blood flow[47] are impaired in obesity, and this is associated with reduced nutrient disposal.[48] In our human intervention trial showing that supplementation with DHA-rich tuna oil reduced body fat in overweight and obese adults,[23] we found that the n-3 LCPUFA also improved flow-mediated dilatation. Thus, some of the observed reduction in body fat may have resulted from the improved vasodilator function increasing blood flow and nutrient delivery to skeletal muscle during exercise. It is possible that impaired perfusion of skeletal muscle in obese subjects may limit their capacity for exercise-induced increases in fat metabolism, thus further exacerbating their condition. No studies have directly addressed this hypothesis, but given the relationships between obesity and skeletal muscle blood flow, and skeletal muscle blood flow and nutrient disposal, additional studies seem warranted.

CONCLUSION

There is mixed evidence from animal and human studies for effects of n-3 LCPUFA on body fat, with a number of studies suggesting that increased consumption of n-3 LCPUFA, particularly DHA, may help to reduce body fat. Such reductions in body fat could result from a range of mechanisms, including appetite-suppressing effects, changes in the expression of genes in metabolic pathways in a range of tissues which shift metabolism toward increased fat oxidation and energy expenditure and reduced fat deposition or, alternatively, from enhanced delivery of nutrients to skeletal muscle. There is also some evidence that n-3 LCPUFA might increase anabolic pathways in skeletal muscle leading to the maintenance of a greater lean tissue mass and increased metabolic rate. Additional well-controlled human trials are required to confirm the importance of n-3 LCPUFA intake for the attainment and maintenance of healthy weight and body composition to determine its efficacy for body fat reduction in overweight individuals and to establish the underlying mechanisms of action.

REFERENCES

1. He K, Rimm E, Merchant A, et al: Fish consumption and risk of stroke in men. JAMA 2002; 288(24): 3130–6.
2. Iso H, Rexrode KM, Stampfer MJ, et al: Intake of fish and omega-3 fatty acids and risk of stroke in women. JAMA 2001; 285(3): 304–12.
3. Micaleff M, Munro I, Phang M, Garg M: Plasma *n*-3 polyunsaturated fatty acids are negatively associated with obesity. Br J Nutr 2009; 102: 1370–4.
4. Klein-Platat C, Drai J, Oujaa M, Schlienger J-L, Simon C: Plasma fatty acid composition is associated with the metabolic syndrome and low-grade inflammation in overweight adolescents. Am J Clin Nutr 2005; 82: 1178–84.
5. Karlsson M, Marild S, Brandberg J, Lonn L, Friberg P, Strandvik B: Serum phospholipid fatty acids, adipose tissue, and metabolic markers in obese adolescents. Obesity 2006; 14(11): 1931–9.
6. Brown A, Pang E, Roberts D: Persistent changes in the fatty acid composition of erythrocyte membranes after moderate intake of n-3 polyunsaturated fatty acids: study design implications. Am J Clin Nutr 1991; 54(4): 668–73.
7. Howe P, Coates A, Murphy K, Pettman T, Milte C, Buckley J: Higher erythrocyte LCn-3 PUFA content is associated with a healthier body composition. Australas Med J 2014; 1(1): 60.
8. Hainault I, Carlotti M, Hajduch E, Guichard C, Lavau M: Fish oil in a high lard diet prevents obesity, hyperlipidemia, and adipocyte insulin resistance in rats. Ann N Y Acad Sci 1993; 683: 98–101.
9. Belzung F, Raclot T, Groscolas R: Fish oil n-3 fatty acids selectively limit the hypertrophy of abdominal fat depots in growing rats fed high-fat diets. Am J Physiol 1993; 264: R1111-8.
10. Baillie R, Takada R, Nakamura M, Clarke S: Coordinate induction of peroxisomal acyl-CoA oxidase and UCP-3 by dietary fish oil: a mechanism for decreased body fat deposition. Prostaglandins Leukot Essent Fatty Acids 1999; 60(5–6): 351–6.
11. Takahashi Y, Ide T: Dietary n-3 fatty acids affect mRNA level of brown adipose tissue uncoupling protein 1, and white adipose tissue leptin and glucose transporter 4 in the rat. Br J Nutr 2008; 84(2): 175–84.
12. Ruzickova J, Rossmeisl M, Prazak T, et al: Omega-3 PUFA of marine origin limit diet-induced obesity in mice by reducing cellularity of adipose tissue. Lipids 2004; 39(12): 1177–85.
13. Pérez-Matute P, Pérez-Echarri N, Martínez JA, Marti A, Moreno-Aliaga MJ: Eicosapentaenoic acid actions on adiposity and insulin resistance in control and high-fat-fed rats: role of apoptosis, adiponectin and tumour necrosis factor-α. Br J Nutr 2007; 97: 389–98.
14. Huang XF, Xin X, McLennan P, Storlien L: Role of fat amount and type in ameliorating diet-induced obesity: insights at the level of hypothalamic arcuate nucleus leptin receptor, neuropeptide Y and pro-opiomelanocortin mRNA expression. Diab Obes Metab 2004; 6(1): 35–44.
15. 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 Obesity 1997; 21: 637–43.
16. Groh-Wargo S, Jacobs J, Auestad N, O'Connor DL, Moore JJ, Lerner E: Body composition in preterm infants who are fed long-chain polyunsaturated fatty acids: a prospective, randomized, controlled trial. Pediatr Res 2005; 57(5): 712–8.
17. Kabir M, Skurnik G, Naour N, et al: Treatment for 2 mo with n-3 polyunsaturated fatty acids reduces adiposity and some atherogenic factors but does not improve insulin sensitivity in women with type 2 diabetes: a randomized controlled study. Am J Clin Nutr 2007; 86: 1670–9.
18. Todoric J, Löffler M, Huber J, et al: Adipose tissue inflammation induced by high-fat diet in obese diabetic mice is prevented by n-3 polyunsaturated fatty acids. Diabetologia 2006; 49: 2109–19.
19. Crochemore IC, Souza AF, de Souza AC, Rosado EL: Omega-3 polyunsaturated fatty acid supplementation does not influence body composition, insulin resistance, and lipemia in women with type 2 diabetes and obesity. Nutr Clin Pract 2012; 27(4): 553–60.
20. Rafraf M, Mohammadi E, Asghari-Jafarabadi M, Farzadi L: Omega-3 fatty acids improve glucose metabolism without effects on obesity values and serum visfatin levels in women with polycystic ovary syndrome. J Am Coll Nutr 2012; 31(5): 361–8.
21. Warner J, Ullrich I, Albrink M, Yeater R: Combined effects of aerobic exercise and omega-3 fatty acids in hyperlipidemic persons. Med Sci Sports Exerc 1989; 21(5): 498–505.
22. Brilla L, Landerholm T: Effect of fish oil supplementation and exercise on serum lipids and aerobic fitness. J Sports Med Phys Fitness 1990; 30: 173–80.
23. Hill A, Buckley J, Murphy K, Howe P: Combining fish oil supplementation with regular aerobic exercise improves body composition and cardiovascular risk factors. Am J Clin Nutr 2007; 85: 1267–74.
24. Wing S, Goldberg A: Glucocorticoids activate the ATP-ubiquitin-dependent proteolytic system in skeletal muscle during fasting. Am J Physiol 1993; 264: E668–76.
25. Whitehouse A, Tisdale M: Downregulation of ubiquitin-dependent proteolysis by eicosapentaenoic acid in acute starvation. Biochem Biophys Res Comm 2001; 285: 598–602.
26. Gingras A, White P, Chouinard P, et al: Long-chain omega-3 fatty acids regulate bovine whole-body protein metabolism by promoting muscle insulin signalling to the Akt–mTOR–S6K1 pathway and insulin sensitivity. J Physiol 2007; 579: 269–84.
27. Fontani G, Corradeschi F, Felici A, et al: Blood profiles, body fat and mood state in healthy subjects on different diets supplemented with Omega-3 polyunsaturated fatty acids. Eur J Clin Invest 2005; 35(8): 499–507.
28. Krebs J, Browning L, McLean N, et al: Additive benefits of long-chain n-3 polyunsaturated fatty acids and weight-loss in the management of cardiovascular disease risk in overweight hyperinsulinaemic women. Int J Obesity 2006; 30:1535–44.
29. Thorsdottir I, Tomasson H, Gunnarsdottir I, et al: Randomized trial of weight-loss-diets for young adults varying in fish and fish oil content. Int J Obesity 2007; 31: 1560–6.
30. Parra D, Ramel A, Bandarra N, Kiely M, Martinez J, Thorsdottir I: A diet rich in long chain omega 3 fatty acids modulates satiety in overweight and obese volunteers during weight loss. Appetite 2008; 51: 676–80.
31. Kunešová M, Braunerová R, Hlavatý P, et al: The influence of n-3 polyunsaturated fatty acids and very low calorie diet during a short-term

weight reducing regimen on weight loss and serum fatty acid composition in severely obese women. Physiol Res 2006; 55: 63–72.
32. DeFina LF, Marcoux LG, Devers SM, Cleaver JP, Willis BL: Effects of omega-3 supplementation in combination with diet and exercise on weight loss and body composition. Am J Clin Nutr 2011; 93(2): 455–62.
33. Munro IA, Garg ML: Dietary supplementation with long chain omega-3 polyunsaturated fatty acids and weight loss in obese adults. Obes Res Clin Pract 2013; 7(3): e173–81.
34. Munro IA, Garg ML: Prior supplementation with long chain omega-3 polyunsaturated fatty acids promotes weight loss in obese adults: a double-blinded randomised controlled trial. Food Funct 2013; 4(4): 650–8.
35. Munro IA, Garg ML: Dietary supplementation with n-3 PUFA does not promote weight loss when combined with a very-low-energy diet. Br J Nutr 2012; 108(8): 1466–74.
36. Gray B, Steyn F, Davies PS, Vitetta L: Omega-3 fatty acids: a review of the effects on adiponectin and leptin and potential implications for obesity management. Eur J Clin Nutr 2013; 67(12): 1234–42.
37. Power G, Newsholme E: Dietary fatty acids influence the activity and metabolic control of mitochondrial carnitine palmitoyltransferase I in rat heart and skeletal muscle. J Nutr 1997; 127: 2142–50.
38. Flachs P, Horakova O, Brauner P, et al: Polyunsaturated fatty acids of marine origin upregulate mitochondrial biogenesis and induce β-oxidation in white fat. Diabetologia 2005; 48: 2365–75.
39. Mori T, Kondo H, Hase T, Tokimitsu I, Murase T: Dietary fish oil upregulates intestinal lipid metabolism and reduces body weight gain in C57BL/6J mice. J Nutr 2007; 137(12): 2629–34.
40. Barber E, Sinclair AJ, Cameron-Smith D: Comparative actions of omega-3 fatty acids on in-vitro lipid droplet formation. Prostaglandins Leukot Essent Fatty Acids 2013; 89(5): 359–66.
41. Wyke S, Tisdale M: NF-kB mediates proteolysis-inducing factor induced protein degradation and expression of the ubiquitin–proteasome system in skeletal muscle. Br J Cancer 2005; 92: 711–21.
42. Buckley J, Burgess S, Murphy K, Howe P: DHA-rich fish oil lowers heart rate during submaximal exercise in elite Australian Rules footballers. J Sci Med Sport 2009; 12: 503–7.
43. Peoples G, McLennan P, Howe P, Groeller H: Fish oil reduces heart rate and oxygen consumption during exercise. J Cardiovasc Pharmacol 2008; 52: 540–7.
44. Davison K, Bircher S, Hill A, Coates AM, Howe PR, Buckley JD: Relationships between obesity, cardiorespiratory fitness, and cardiovascular function. J Obes 2010; 2010: 191–253.
45. Hamdy O, Ledbury S, Mullooly C, et al: Lifestyle modification improves endothelial function in obese subjects with the insulin resistance syndrome. Diab Care 2003; 26(7): 2119–25.
46. Parikh N, Keyes M, Larson M, et al: Visceral and subcutaneous adiposity and brachial artery vasodilator function. Obesity 2009; 17(11): 2054–9.
47. Hodnett B, Hester R: Regulation of muscle blood flow in obesity. Microcirculation 2007; 14: 273–88.
48. Clerk L, Vincent M, Jahn L, Liu Z, Lindner J, Barrett E: Obesity blunts insulin-mediated microvascular recruitment in human forearm muscle. Diabetes 2006; 55:1436–42.