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REVIEW

Indications for Omega-3 Long Chain Polyunsaturated Fatty Acid in the Prevention and Treatment of Cardiovascular Disease

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Background The National Heart Foundation of Australia (NHFA) 2008 review on omega-3 long-chain polyunsaturated fatty acids (LCPUFA) made recommendations with respect to supplementation for primary and secondary prevention of cardiovascular disease. Since then, new findings have been published regarding the relationship between omega-3 polyunsaturated fatty acids, including supplementation, and cardiovascular health.

Methods A literature search was undertaken in PubMed and Medline, for literature published between January 1, 2007 and August 31, 2013.

Results and Conclusions A total of eight research questions were developed and, using the National Health and Medical Research Council's evidence assessment framework, conclusions were made in relation to dietary intake of fish and omega-3 LCPUFA for cardiovascular health.

In the evidence published since 2007, this summary of evidence concludes that dietary intake of fish was found to be mostly consistent with respect to protection from heart disease and stroke. Higher fish intake was associated with lower incident rates of heart failure in addition to lower sudden cardiac death, stroke and myocardial infarction.

In relation to omega-3 LCPUFA supplementation, neither a beneficial nor adverse effect was demonstrated in primary or secondary prevention of coronary heart disease (CHD). Although the evidence continues to be positive for the role of omega-3 LCPUFA in the treatment of hypertriglyceridaemia and a modest positive benefit in heart failure. No further evidence was found to support the consumption of 2 g alpha-linolenic acid (ALA)/day over the current Australian guidelines for 1 g/day.

Keywords Fish oil • Omega-3 polyunsaturated fatty acid supplements • Fish consumption • Cardiovascular disease • Recent evidence.

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Introduction

In 2008, the National Heart Foundation of Australia (NHFA) position statement [1] on omega-3 long-chain polyunsaturated fatty acids (LCPUFA) recommended Australian adults should consume 500 mg combined docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) and 2 g alpha-linolenic acid (ALA) per day for primary prevention of cardiovascular disease, 1000 mg EPA/DHA and 2 g ALA per day for secondary prevention, and 1-4 g EPA/DHA per day for treatment of hypertriglyceridaemia.

In recent years, intervention trials with omega-3 LCPUFA supplements have started to trend towards no effect at least with respect to primary and secondary prevention of coronary heart disease (CHD). These outcomes have been perplexing, based on the established benefits of omega-3 LCPUFA on potential mechanistic cardiovascular pathways.

In view of the addition of new research to the evidence base, and questions of uncertainty regarding dietary and/or supplementation of omega-3 LCPUFA for heart health, the National Heart Foundation of Australia implemented a review of the literature published since the 2008 Position Statement. Major publications are evaluated in the current paper and lend support to the Foundation's changed recommendations in 2014.

This 2014 summary updates the evidence for omega-3 LCPUFA, and provides guidance for health professionals on dietary intake of fish and omega-3 LCPUFA supplements.

Methodology

The literature search was undertaken in PubMed and Medline, for literature published between January 1, 2007 and August 31, 2013. The literature searches used key search words including but not limited to ''exp Eicosapentaenoic Acid/ or exp Fatty Acids, Omega-3/ or exp Docosahexaenoic Acids/'' AND ''exp Platelet Aggregation/ or exp Endothelium, Vascular/'', ''exp Stroke/'', ''exp Arrhythmias, Cardiac/ OR exp Atrial Fibrillation/ OR exp Tachycardia, Ventricular/'', ''exp Triglycerides/'', ''myocardial infarction'', ''coronary event'', ''coronary disease'', ''coronary heart disease'', ''heart failure'', and ''exp Cardiovascular Diseases/ and exp alpha-Linolenic Acid/''. This report deals with those aspects where adequate evidence was available for grading. Searches were limited to clinical trials, cohorts, comparative studies, meta-analyses, multicentre studies, randomised controlled trials (RCT) or systematic reviews. Animal studies were excluded as were studies with inappropriate study design (i.e. cross-sectional survey or narrative review) or small-sized or underpowered. Literature published in 2007 was cross-checked with the 2008 Position Paper to avoid duplication. A desktop review was undertaken to identify and review clinical and dietary guidelines relevant to cardiovascular health, and for guidelines relevant for other conditions.

The evidence statements and the recommendations made in this consensus statement have been graded according to National Health and Medical Research Council guidelines [2] (Table 1). Assessing the evidence for omega-3 LCPUFA was based on intervention studies with supplements of one or two major fatty acids or on observational studies that documented consumption of fish. The 'intervention' hierarchy of evidence was used and preferred as the key criterion of efficacy. Using this hierarchy of evidence, the randomised controlled trials (RCTs)/meta-analyses outcomes are judged to be a higher level than prospective cohort studies (no higher than C) by definition although alternative higher level gradings have been recently put forward by the NHMRC.

Table 1 **National Health and Medical Research Council: Evidence Hierarchy and Assessment Matrix[1].**

Level of evidence	**Study design (Intervention)**
I	A systematic review of Level II studies
II	Evidence obtained from at least one properly designed RCT.
III-1	Evidence obtained from well-designed, pseudo RCTs (alternate allocation or some other method).
III-2	Evidence obtained from comparative studies with concurrent controls and allocation, not randomised cohort studies, case-control studies or interrupted time series with a control group.
III-3	Evidence obtained from comparative studies with historical control, two or more single-arm studies, or interrupted time series without a parallel control group.
IV	Evidence obtained from case series, either post-test or pre-test and post-test.
Grade of Recommendation	**Description**
A	Body of evidence can be trusted to guide practice
B	Body of evidence can be trusted to guide practice in most situations
C	Body of evidence provides some support for recommendation(s) but care should be taken in its application
D	Body of evidence is weak and recommendation must be applied with caution

[1] Adapted from source: National Health and Medical Research Council. NHMRC additional levels of evidence and grades for recommendations for developers of guidelines. 2009.

Results and Discussion (Table 2)

Primary Prevention of Coronary Heart Disease

The search strategy identified 10 sources concerning omega-3 LCPUFA intake in the primary prevention of coronary heart disease (CHD) since 2008. The data include two systematic reviews and meta-analyses [3,4], three adequately powered RCTs [5–7] as well as six prospective cohort studies [8–13].

Are omega-3 LCPUFA supplements effective in the primary prevention of coronary heart disease?

Two Level I studies [3,4] (meta-analyses by Kotwal et al., and Rizos et al.,) and three Level II studies (RCTs) [5–7] were accepted as post-2008 publications. RCTs included the ORIGIN trial of subjects at high risk primarily from dysglycaemia [5]; a further analysis of the Japanese JELIS trial [6] in a hypercholesterolaemic population; and the Risk and Prevention Study [7] comprising many diabetic subjects. In these studies the dose of EPA or EPA/DHA was 1-2 g and duration was sufficiently long to accumulate large numbers of outcome events. The two meta-analyses [3,4] found no statistical evidence of effect on myocardial infarction, but a beneficial effect was found for cardiovascular death prevention (in Rizos analysis: Relative Risk [RR], 0.91; 95% CI 0.85-0.98). The JELIS analysis suggested an inverse association for higher plasma levels of EPA (a biomarker of compliance) and coronary events (HR 0.83; p=0.049). Neither ORIGIN nor the Risk and Prevention study found significant beneficial effects of supplementation.

> ***The recommendation from these data is that there has been no further evidence to recommend supplementation with omega-3 LCPUFA for primary prevention (Grade C).***

Is the reported consumption of omega-3 LCPUFA from fish, or dietary patterns high in omega-3 LCPUFA (measured through plasma or red blood cell LCPUFA) associated with lower incidence of coronary heart disease events in primary prevention?

Prospective cohort studies comprised medium to long-term evaluation of CVD/CHD outcomes in populations whose consumption of fish had been assessed by standard dietary questionnaires or by biomarkers of fish intake (plasma EPA and DHA). This is a clear distinction from the previous studies in which supplements were taken. Population and supplemental clinical controlled trials all are confounded by not only background intake of fish and other foods but also how the flesh is cooked and eaten and genetic factors which influence plasma and corresponding tissue levels. Six studies [8–13] of acceptable quality and low to moderate bias were identified as post-2008 publications. All were classified as having a Level III evidence base, two were systematic reviews and meta-analyses [11,13] of prospective cohorts and so were considered as having the highest category at Level III. Four others were judged as Level III-1 [8–10,12]. The Zheng et al. [13] meta-analysis of 17 cohorts with 315,812 participants and an average follow-up of 15.9 years, concluded that compared to no intake, both low (one serving/week) and moderate (two to four servings/week) fish consumption significantly prevented CHD mortality (0.84, 95%CI 0.75-0.95; 0.79, 95%CI 0.67-0.92 respectively). Musa-Veloso et al. [11] also concluded a benefit with >250 mg omega-3 LCPUFA from fish reducing the risk of sudden cardiac death by 35%. The four Level III-1 studies uniformly demonstrated inverse associations with sudden cardiac death. On aggregate, these studies provided a sufficiently large number of events to warrant a different conclusion from the Level I and Level II studies in which supplements were used. The daily intake of omega-3 LCPUFA above which positive outcome incidence diminished varied between 250 mg to 500 mg daily [11,13].

A recurring issue has been the suggestion of an adverse interaction between the consumption of n-6 fatty acid (primarily linoleic acid) and omega-3 LCPUFA which has been adequately addressed in a recent evaluation of both fatty acids in a large long-term prospective cohort study [14]. Although the primary objective was to demonstrate the potential benefit of n-6 fatty acids, evaluated by phospholipid fatty acid analyses, the study of 2792 elderly participants showed substantial reductions in both cardiovascular and all-cause mortality when the two sources of polyunsaturated fatty acids were combined. Comparing participants with the

Table 2 Evidence statements regarding omega-3 long-chain polyunsaturated fatty acid intake and heart disease.

In the studies published since 2008, there is no further evidence that omega-3 LCPUFA supplementation is beneficial as an intervention for the primary prevention of coronary heart disease [3–7].	I	C
In the studies published since 2008, there is good evidence that increased consumption of fish or dietary patterns with omega-3 LCPUFA are associated with the primary prevention of coronary heart disease [8–12].	III	**B**
In studies published since 2008, there is no further evidence of a benefit from omega-3 LCPUFA supplementation in the secondary prevention of coronary heart disease [3,4,15,18,19,26].	I	C
There is insufficient evidence published since the 2008 paper to assess the relationship between fish consumption or dietary patterns with omega-3 PUFA in secondary prevention of coronary heart disease.	n/a	n/a
There is some evidence for a modest benefit from omega-3 polyunsaturated fatty acid supplementation in addition to standard therapy, in heart failure [20,23–25,28].	**II**	**C**
There is further evidence for omega-3 LCPUFA supplementation in the treatment of hypertriglyceridaemia [29,30].	**I**	**A**
There is no further evidence that intervention with alpha-linolenic acid reduces risk of coronary heart disease outcomes.	n/a	n/a

highest versus the lowest levels of phospholipid fatty acids led to a 64% lower cardiovascular mortality and a 54% lower total mortality, with each fatty acid class contributing independently to the overall benefit.

The recommendation from these studies supported greater consumption of fish (including oily fish) (Grade B) and thus did not differ from the Heart Foundation's 2008 advice.

Secondary Prevention of Coronary Heart Disease

The search strategy identified 23 sources since 2008 regarding omega-3 LCPUFA intake in the secondary prevention of coronary heart disease. The data that met inclusion criteria include five systematic reviews and meta-analyses [3,4,15–17], three adequately powered RCTs [18–20] as well as two prospective cohort studies [21,22] and three prospective cohort studies specific to heart failure [23–25].

There were more Level I and Level II studies than for primary prevention. These refer to studies that investigated use of supplemental omega-3 LCPUFA. Level III studies based on prospective cohort studies were few in number. Several studies included both primary and secondary category patients and hence appear in both sections.

Are omega-3 LCPUFA supplements an effective intervention for the secondary prevention of coronary heart disease?

Five Level 1 meta-analyses were included: Kotwal et al.; Rizos et al.; Kwak et al.; Zhao et al., and Marik et al. Three Level II RCTs included the OMEGA study [19], Alpha to Omega study [18], and GISSI Heart Failure (GISSI-HF) trial [20]. Two RCTS [19,20] provided 1 g supplements of EPA/DHA, one for only one year (OMEGA and GISSI-HF), while the Alpha-Omega trial provided approximately 400 mg EPA/DHA which equates favourably with dietary guideline recommendations [18]. The two meta-analyses published prior to 2010 [16,17] provided evidence of benefit in patients with existing CHD, but the meta-analyses published after 2010 did not [3,4,15]. The OMEGA and the Alpha to Omega RCTs did not find evidence of a beneficial effect.

As was the case for primary prevention, the evidence reviewed regarding supplementation for secondary prevention failed to provide further support for the use of omega-3 LCPUFA supplements to prevent recurrent CHD. (Grade C). However a case was made out for their use as an adjunct to heart failure therapy (see 1.5).

Is the reported consumption of fish, or dietary patterns high in omega-3 LCPUFA (measured through plasma or red blood cell LCPUFA) associated with a lower incidence of coronary heart disease in patients with existing CHD (i.e. secondary prevention)?

Evidence from prospective cohort studies of subjects with existing CVD/CHD was classified as Level III. Since there were only two applicable studies [21,22], *no clear new recommendation is possible other than to support the consumption of fish including oily fish on general principles as concluded in the 2008 review.*

Are omega-3 LCPUFA supplements effective in the prevention or treatment of heart failure?

There has been one large outcome trial (GISSI-HF) and a number of smaller studies that have assessed omega-3 LCPUFA intake on left ventricular function [26]. In the GISSI-HF trial 7,000 patients with functional Class II to IV heart failure were randomised to 1 g/day or placebo. Over a 3.9-year median follow-up, supplementation resulted in an absolute 9% reduction in mortality or admission to hospital (p=0.04). Benefit was greater in elderly and diabetic patients (sub-groups with greater absolute risk).

The GISSI-P study [27] was the first large randomised but unblinded trial that demonstrated that omega-3 LCPUFA supplementation following an acute myocardial infarction reduced mortality. Later analysis of this study demonstrated that the reduction in sudden cardiac deaths (SCD) was greater in those patients with impaired left ventricular function (trend p=0.01) [28]. Comparing SCD for patients with ejection fractions <40% with those >50%, there was a four-fold greater benefit with supplementation (58% reduction vs 11% p=0.0003).

Results from GISSI-HF, and later analysis of GISSI-P, provide modest support for 1 g of omega-3 LCPUFA (Grade C) in addition to standard therapy in patients with heart failure.

Is the reported consumption of fish, or dietary patterns high in omega-3 LCPUFA (measured through plasma or red blood cell LCPUFA) associated with lower incidence of heart failure?

A meta-analysis of seven prospective cohort studies that involved 176,441 subjects with 5,480 incident cases of heart failure, showed that high fish intake was protective against developing HF [23]. The Cardiovascular Health Study examined 4,738 US adults older than 65 but free of HF at baseline [24]. After 12 years, 955 patients developed heart failure. There was an inverse relationship between fish intake and incidence of heart failure. The highest quintile had a 32% lower risk compared to those who consumed fish less than or equal to once a month (p trend 0.009). A 14.3-year follow-up from The Atherosclerosis Risk in Communities (ARIC) study showed plasma phospholipid omega-3 LCPUFA (especially EPA) at baseline inversely correlated with heart failure in women but not in men (P<0.001) [25].

These observational data are supportive of a modest inverse association between fish consumption and heart failure (insufficient studies for grading).

Hypertriglyceridaemia

Are omega-3 LCPUFA supplements an effective intervention for lowering plasma triglycerides in hypertriglyceridaemic patients?

The search strategy identified 63 sources of clinical information concerning the effect of omega-3 LCPUFA on plasma

triglyceride levels that have been published since 2008. The data that met inclusion criteria include two meta-analyses [29,30] and numerous smaller RCTs which report the plasma triglyceride response.

The meta-analysis by Reiner et al. highlights the relative safety of omega-3 LCPUFA as a triglyceride-lowering therapy [30], with the caveat that they may augment the anti-platelet effects of combination therapy with aspirin and clopidogrel. Another meta-analysis restricted to patients with diabetes noted that triglyceride reduction may be accompanied by an increase in LDL-cholesterol [29]. Meta-analyses and small individual studies extend the clinical settings in which omega-3 LCPUFA reduce hypertriglyceridaemia by including groups of patients with chronic renal disease [31], HIV receiving HAART [32], metabolic syndrome [33], dysglycaemia [5] and other conditions.

It is important to emphasise that omega-3 LCPUFA are not an effective therapy for the control of LDL-cholesterol [34]. Hypertriglyceridaemia represents a situation in which the relationship between LDL-cholesterol and CHD risk is moderately confounded by changes in LDL-composition, and it is possible that the difference between total and HDL-cholesterol, the so-called ''non-HDL'' cholesterol, may represent a more accurate estimate of CHD risk. Omega-3 LCPUFAs have been shown to favourably reduce non-HDL cholesterol [35].

These data confirm omega-3 LCPUFA as a means for lowering plasma triglyceride levels (Grade A).

Alpha-Linolenic Acid (ALA)

Alpha-linolenic acid (ALA) is derived from plants and is found mainly in fats and oils, such as canola oil and soybean oil, and in linseeds and walnuts. ALA is an essential fatty acid in humans that must be supplied in the diet. The search strategy identified 12 sources of clinical information concerning the effects of ALA on cardiovascular disease that have been published since 2008. The data that met inclusion criteria include two meta-analyses [36,37], one RCT [18] and two cohort studies [38,39]. *The 2008 position statement did not find high-quality conclusive data to change the Heart Foundation's 1999 recommendation for 2 g ALA in secondary prevention of CHD.*

Is the intervention with the plant omega-3 fatty acid (alpha-linolenic acid) or its consumption effective in the prevention of coronary heart disease?

Pan et al. conducted a meta-analysis in 2012 and examined 27 dietary and biomarker studies relating ALA to CVD events [36]. Overall there was a 14% higher incidence in total events from the lowest to the highest tertile [36] that was significant for the dietary assessment studies but not for the biomarker assessment studies. However, there was significant heterogeneity in both types of studies. CHD mortality was the only factor that was significantly lower (20%, six cohort studies). Each 1 g/day intake of ALA was associated with a 10% lower risk of CHD death. The authors noted that there are no randomised, placebo controlled trials evaluating the effects of ALA on primary prevention of CVD events. In 4837 post-MI patients, there was no statistically significant relationship between supplementation with 2 g ALA and control in a randomised, placebo-controlled trial [18]. Neither ALA intake nor phospholipid ALA level was associated with incident atrial fibrillation [38] or heart failure [39]. However the authors noted that plasma ALA may not be a good biomarker of dietary ALA intake as ALA is rapidly oxidised after consumption. High ALA diets do not lower plasma triglycerides [40] but there have been few studies in hyperlipaemic subjects. A review that included 28 flaxseed interventions found no effects of flaxseed oil [37] on HDL-cholesterol and triglycerides, and only a marginal effect on total and LDL-cholesterol that was not consistent across all subgroups. High ALA intake likely reflects intake of soybean oil, canola oil and walnuts, so other components of these foods beside ALA may confer benefit.

These data provide no evidence to support an intervention with 2 g ALA per day, although higher dietary ALA intake is associated with reduced risk of CHD death in the setting of primary prevention. The evidence supports including sources of ALA in the diet but not for supplementation.

Other Relevant Cardiovascular Outcomes and Mechanisms of Action

In reviewing the evidence, the role of omega-3 LCPUFA and fish consumption on other cardiovascular outcomes was assessed, including atrial fibrillation and other arrhythmias, blood pressure and stroke. While not assessed using the NHMRC process, a brief narrative review of these relationships is outlined below.

Does Intake of Omega-3 LCPUFA Alter the Risk or Incidence of Atrial Fibrillation?

There have been a number of epidemiological and randomised control trials assessing the efficacy of omega-3 LCPUFA for the prevention of atrial fibrillation (AF). The results have been mixed, either showing no benefit or some benefit [41]. In the Kuopio Ischaemic Heart Disease Study [42], the highest blood level of DHA plus EPA and DPA was associated with a 50% lower risk of developing AF.

In the coronary post-bypass surgery studies, recent meta-analyses of post-operative AF from eight trials that included 2687 patients reported different conclusions. Costanzo et al. showed that supplementation with omega-3 LCPUFA reduced AF by 25% (95% CI, 0.59-1.00 P=0.05) [43]. In contrast Mariani et al. reported a non-significant 14% reduction (95% CI, 0.71-1.04) in AF after omega-3 LCPUFA [44].

Supplementation with omega-3 LCPUFA prior to elective cardioversion of AF has shown no overall benefit [45].

Addressing the possible inadequacy of omega-3 LCPUFA supplementation, a recent randomised placebo-controlled trial of high dose (4 g daily for an average of 271 days) failed to reduce AF recurrence in 337 patients with symptomatic persistent or paroxysmal AF not receiving conventional anti-arrhythmic therapy [46].

Do Omega-3 LCPUFA Alter the Risk or Incidence of High Blood Pressure?

There is substantial evidence from RCTs that omega-3 LCPUFA reduce blood pressure (BP) with a greater effect in hypertensive patients than in those with high-normal BP [47]. The dose required to achieve a BP reduction is likely to be at least 3-4 g/day. Three meta-analyses [48–50] have shown that omega-3 LCPUFA reduce BP with the greatest effect in hypertensive patients (-3.4 to -5.5 mmHg systolic BP and -2.0 to -3.5 mmHg diastolic BP). Dokholyan et al. [51] also suggested that greater than 3 g/day omega-3 LCPUFA is required to reduce BP in patients with high-normal diastolic BP or stage 1 hypertension.

Randomised controlled studies have shown the BP-lowering effects of omega-3 LCPUFA are modest and potentiated by lifestyle changes such as weight loss [52], sodium restriction [53] and antihypertensive medication [54]. Mori et al. [55] showed that in overweight, mildly-hypercholesterolaemic patients, 4 g/day of encapsulated DHA, but not EPA, reduced 24-hour BP by -5.8/-3.3 mmHg. Thus, there is evidence in hypertensive patients that current therapy can be complemented with omega-3 LCPUFA supplementation, although more research is required before making firm recommendations.

Are Omega-3 LCPUFA as Supplements or Consumed in Fish Effective in Preventing Ischaemic Stroke? Do they Affect the Risk of Haemorrhagic Stroke?

Mozaffarian et al. (2013) reported in the Cardiovascular Health study that total plasma phospholipid omega-3 LCPUFA were inversely related to ischaemic stroke risk (p=0.043) with a 37% reduction in the highest versus the lowest quintile, but there was no significant effect on haemorrhagic stroke (p=0.86) [10]. DHA was most strongly associated with reduction in ischaemic stroke and DPA with reduction in stroke death. In the same study, the highest quartile of total omega-3 LCPUFA intake (DHA in particular) associated with 40% fewer prevalent subclinical infarcts on first MRI examination (p=0.001) [56]. Larsson et al., in a meta-analysis of fish consumption and stroke in 15 prospective studies, showed that an increment of three servings of fish/week was associated with a 6% lower incidence of total stroke [57]. In nine studies in which stroke was a subset there was a 10% lower risk of ischaemic stroke and a non-significant 10% lower risk of haemorrhagic stroke [57]. Interventions with fish oil supplementation have not demonstrated any reduction in stroke [4]. Thus, the evidence for stroke prevention is stronger for fish consumption than for omega-3 LCPUFA supplements.

Mechanisms Underlying The Effects Of Omega-3 LCPUFA On Cardiovascular Risk Factors

Omega-3 LCPUFA influence a range of physiological and biochemical functions in animal and clinical studies that would be expected to translate potentially into fewer adverse CVD outcomes. Such associations have been more difficult to demonstrate in RCTs of CVD endpoints.

Omega-3 LCPUFA have been shown to alter plasma lipoproteins, vascular function, inflammatory markers, cardiac function and thrombogenicity. A multiplicity of beneficial molecular and biochemical mechanisms have been demonstrated at a cellular level and in animal models.

Effect on Plasma Lipoproteins

Hepatic production of the key triglyceride-rich lipoprotein, very low density lipoprotein (VLDL) is reduced due to suppression of synthesis of apolipoprotein B and triglycerides. Oxidation of fatty acids in liver is accelerated reducing their incorporation into triglycerides and pathways of triglyceride synthesis are suppressed. The composition of LDL particles and the inverse relationship between circulating triglycerides and HDL levels are due in part to the transporter cholesteryl ester transfer protein (CETP) that mediates exchange of lipids between lipoproteins enriching HDL with cholesterol. Suppression of CETP activity by omega-3 LCPUFA is partly responsible for the modest increase in HDL cholesterol [58]. LDL-cholesterol concentration may rise modestly with fish oil supplementation [59].

Vascular Function

Vascular dysfunction associates with increased CVD risk. Stiffness of large arteries due to aging or disease associates with CVD events. The integrity and health of the vascular endothelium is a determinant of normal circulatory function. Endothelial dysfunction, through increased inflammatory and thrombogenic molecular changes, has also been associated with higher CVD event rates. Importantly both arterial stiffness and endothelial dysfunction are ameliorated through omega-3 LCPUFA consumption [60–62].

Omega-3 fatty acids improve vascular function in healthy individuals [63], and in hypercholesterolaemic [64] and type 2 diabetic [65] patients. Mori et al. [61] showed that in dyslipidaemic patients DHA and not EPA, improved endothelial and smooth muscle function and also reduced vasoconstrictor responses.

Fish oil [66] and both EPA and DHA supplementation [62] reduce arterial stiffness. In a meta-analysis that included 10 randomised controlled trials, Pase et al. [67] showed that omega-3 fatty acids significantly improved both pulse wave velocity and arterial compliance.

Omega-3 fatty acids reduce heart-rate [68] suggesting a significant cardiac component associated with the antihypertensive effects, possibly mediated by effects on cardiac myocytes, autonomic nerve function or beta-adrenoreceptor activity. In a meta-analysis, Mozaffarian et al. [68] showed that omega-3 LCPUFA reduce heart-rate by -1.6 bpm, with a greater reduction in individuals with a heart rate >69 bpm (-2.5 bpm) and in those studies of longer than 12 weeks' duration (-2.5 bpm).

Xin et al. [69] in a meta-analysis of 15 randomised controlled trials showed that short-term omega-3 LCPUFA supplementation favourably affects the frequency of heart rate variability through the enhancement of vagal tone.

Inflammation, Thrombogenicity and Oxidisability

These three processes, which are partly inter-related at molecular levels, are key mediators of atherosclerosis as well as of vascular dysfunction. Studies that have shown reduction in inflammatory markers such as NFK-beta, IL6 and TNF-alpha have generally used more than 3000 mg/day. In humans the concentrations of circulating biomarkers indicating inflammation and thrombogenic activity have been reported to be reduced with omega-3 LCPUFA consumption [70]. Less certain is the clinical application of these findings, at least at dosages commonly consumed. Conflicting reports regarding the concentrations of inflammatory and thrombogenic (haemostatic) biomarkers in populations of fish eaters or after interventions with LCPUFA demonstrate the difficulty of reconciling clinical observations with physiological data. However these effects may contribute to improved endothelial function and possibly to the pathology of atheromatous plaques.

Increased oxidative burden, a theoretical disadvantage of the multiple double bonds in omega-3 LCPUFA, has not been demonstrable clinically [71,72].

Do Different Omega-3 LCPUFA (e.g. EPA, DHA or DPA) Confer Different Outcomes?

There are no data to suggest a differential benefit from EPA versus DHA on clinical outcomes. The GISSI-HF intervention [20] which showed benefit in heart failure was both EPA and DHA rich, while the JELIS intervention [6] which showed benefit for sudden cardiac death was with EPA only. Similarly the neutral studies contained comparable amounts of EPA and DHA or a modest preponderance of EPA. A recent meta-analysis of 21 RCTs of the effects of EPA vs DHA on serum lipids found both reduced triglycerides [73].

Docosapentaenoic acid (DPA) is not a component of fish or fish oil but occurs in meat and is an elongation product of EPA and increases when EPA is consumed. In some studies circulating DPA levels were associated with protection for heart failure [74] and predictive of a decrease in non-cardiovascular mortality [10]. No DPA dietary studies were identified for assessment in this review.

Are Omega-3 LCPUFA Best Consumed as Triglycerides, Phospholipids or Ethyl-esters?

The majority of research to date has used EPA/DHA in the form of ethyl-esters. EPA/DHA in phospholipid form is a little more completely absorbed than in triglyceride form. Krill oil is a phospholipid-rich and free fatty acid rich form of fish oil. Krill oil reduces triglycerides to the same degree as fish oil [75] and both increase plasma EPA and DHA to a similar extent for the same intake of EPA/DHA [76]. There is currently insufficient evidence to support claims of greater efficacy of krill oil.

Individual differences in response to a fixed dose of fish oil can vary considerably. After 28 days of supplementation with 4 g of EPA / DHA, plasma EPA levels were increased by 1.1-6.36% and DHA by 3.4-6.3% [77]. Poor therapeutic responders are likely to have diluted the benefit of omega-3 LCPUFA supplementation on clinical outcomes.

National and International Guidelines For Omega-3 LCPUFA & Fish

In Australia, long-chain omega-3 LCPUFA are listed with the Therapeutic Goods Administration (TGA) for human consumption [78]. Only the ethyl-ester form is registered by the TGA for secondary prevention following an acute myocardial infarction and for hypertriglyceridaemia. The Pharmaceutical Benefits Advisory Committee (PBAC) in 2010 did not approve omega-3 LCPUFA ethyl-esters for reimbursement through the Pharmaceutical Benefits Scheme (PBS) for post-infarction management or hypertriglyceridaemia [79].

In terms of dietary intake, the National Health and Medical Research Council (NHMRC) set nutrient reference values (NRVs) in 2006 for omega-3 LCPUFA recommending both an adequate intake to avoid deficiency and a suggested dietary target to prevent chronic disease [80]. The statutory food agency - Food Standards Australia New Zealand (FSANZ) also supports the consumption of omega-3 LCPUFA by allowing general-level health claims for heart health on commercially available food products [81]. However, the recent Australian Dietary Guidelines [82], while acknowledging that the protective effect of fish consumption is most likely mediated through omega-3 LCPUFA, have not specifically recommended omega-3 LCPUFA nor oily fish, a good source of omega-3 LCPUFA. Instead, the Australian Dietary Guidelines recommend at least two serves of (any) fish per week.

Internationally, guidelines published in the last five years have also varied. European guidelines have recommended the consumption of fish at least twice a week, with one of the serves as oily fish [30,83,84]. In 2013, the European Society of Cardiology noted that current recommendations are to increase omega-3 PUFA through fish consumption, rather than from supplements in patients with stable CHD [83]. The European Society of Cardiology recommends omega-3 LCPUFA for the treatment of hypertriglyceridaemia [30]. The current guideline from the European Society is to consider

intervention at and above 1.7mmol/L and that levels above 10mmol/L indicate treating in order to prevent pancreatitis [30].

In contrast, the National Institute of Clinical Excellence (NICE) has recently stepped away from both omega-3 LCPUFA supplements and oily fish recommendations in the secondary prevention of cardiovascular disease [85]. These guidelines, released in late 2013 advise only to consume a Mediterranean-style diet (which includes fish) but do not routinely recommend consumption of oily fish nor omega-3 LCPUFA supplements. In terms of primary prevention, the current NICE guidelines [86] advise consumption of at least two portions of fish, one being oily, per week but do not advise routine recommendation of omega-3 LCPUFA supplements for the primary prevention of CVD.

The Heart Foundation and the Cardiac Society of Australia and New Zealand, in the update on management of chronic heart failure in 2011, recommended that omega-3 LCPUFA should be considered as a second-line agent in people with heart failure who remain symptomatic despite receiving standard therapies with ACE inhibitors and beta-blockers [87]. In line with the findings of this review and the 2011 Heart Foundation heart failure guidelines, the American Heart Association and American College of Cardiology [88] identified omega-3 LCPUFA supplementation as a reasonable adjunctive therapy in patients with heart failure, unless contraindicated.

Recent Controversies

The role and investigation of omega-3 LCPUFA in human health is not limited to cardiovascular outcomes but is not part of this review. However controversy has arisen in relation to their effects on cancer, specifically prostate cancer. Recently, the SELECT trial, a randomised trial which assessed the efficacy of selenium plus or minus vitamin E compared to placebo in the development of new prostate cancer suggested an association with baseline plasma total phospholipid omega-3 LCPUFA and prostate cancer [89]. However, this trial was not adequately powered nor designed to assess intake of omega-3 fatty acids and risk of prostate cancer. 834 men were diagnosed with prostate cancer from a cohort of 35,553. A meta-analysis of case control and cohort studies published in 2010, included 12 case control studies that involved 5777 cases and 12 cohort studies of 445,820 men with 13,924 prostate cancers [90]. In the analysis of case controls, there was no relationship between higher fish intake and prostate cancer, and in the analysis of cohorts fish consumption was associated with 63% reduction of prostate cancer specific mortality.

Limitations

While not considered the 'highest' form of evidence, this review included observational studies. For the purpose of reviewing the evidence since 2008, it was necessary to apply the same inclusion/exclusion criteria to the literature as that used in the 2008 review, which included observational studies of fish consumption and dietary patterns. The consumption of omega-3 LCPUFA by the Australian population is from both fish consumption and supplements. In reviewing the evidence and forming conclusions for fish consumption, the protective effect of fish and marine omega-3 LCPUFA intake must be considered against the likelihood that regular fish consumption may associate with a healthy diet high in vegetables, pulses, and fruit and low in red meat. Further, high fish consumers in Western countries tend to have a higher socioeconomic position and lower prevalence of depressive symptoms – all significant risk factors for CHD [91–93].

Prospective population studies in which the variable under investigation is a food always carry the limitation of known and unknown confounders. As stated above eating fish regularly is sometimes surrogate for healthier dietary patterns that may include reduced intakes of foods considered less healthy. In the publications that have reported benefit from fish consumption the investigators have adjusted the results in multivariate analyses to take into account cardiovascular risk factors including diabetes, alcohol intake, age, socioeconomic and educational status. In at least one study a number of foods likely to influence cardiovascular risk were also accounted for [10]. Saturated fat intake was reported in one study [21] and found to decline across quartiles whereas omega-3 intake increased. Whether adjustment was made is not reported.

Various conventional measures of fish omega-3 consumption were used and in at least one study intake was validated by its strong correlation with plasma phospholipid omega-3 values [21]. Several studies reported more robust indices of habitual fish consumption by measurements of omega-3 long chain polyunsaturated fatty acid levels in adipose tissue [9] or in plasma phospholipid [10].

Finally, observational studies reflect consumption of all types of fish, although oily fish are highlighted. Whether other constituents of fish also have protective properties has intrigued investigators without clear resolution.

Two other issues could not be resolved: Clear distinction among subjects categorised as fitting the primary prevention category from those at high risk for suffering possible cardiovascular disease could not be validated other than to accept the authors' definition, thus there has not been distinction between primary prevention in healthy adults and primary prevention in high risk groups. Secondly, meta-analyses also contained a probable mixture of subjects qualifying for primary and secondary CVD/CHD and were judged on the authors' definitions.

However, the evidence continues to evolve, and the recent neutral findings should be explored in future research to address the significant limitations evident in the literature. Namely, the uncertainty and questions exists around the appropriate fatty acid (crudely EPA vs DHA), the appropriate vehicle to deliver the fatty acid (phospholipid, triglycerides, ethyl esters), the appropriate dosage, exploration of the dose-response, and the appropriate duration of investigation. Answering these questions may elucidate the current unknowns reflected in the evidence.

Conclusions

This summary of evidence concludes that neither beneficial nor adverse effects of omega-3 LCPUFA supplementation were found in primary or secondary prevention of coronary heart disease (CHD), no evidence of harm, and no further evidence was found to support the consumption of 2 g alpha-linolenic acid (ALA)/day over the current Australian guidelines for at least 1 g/day.

However, the evidence continues to be positive for the role of omega-3 LCPUFA in treatment of hypertriglyceridaemia and a modest but positive benefit in heart failure. There is some indication of benefit in atrial fibrillation and hypertension – but further trials are required to support any firm recommendations.

Dietary intake of fish was consistently found to be of benefit for the protection from heart disease and stroke. Higher fish intake was associated with lower incident rates of heart failure in addition to lower sudden cardiac death, stroke and myocardial infarction.

Based on the evidence reviewed in this 2014 paper, and in relation to the existing 2008 evidence and recommendations, a revised recommendation is proposed. The Heart Foundation recommends that all Australians should aim to include two to three serves of fish (including oily fish) per week as part of a heart healthy eating pattern. This amount of fish provides between 250-500 mg per day of combined docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA). Sources of ALA, such as walnuts, flaxseed (linseed), canola and soybean oils should also be included in a healthy eating patterns. Omega-3 LCPUFA supplements can be considered in patients with heart failure in additional to standard therapy [87]. Omega-3 LCPUFA supplements are effective in the treatment of hypertriglyceridaemia.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.hlc.2015.03.020.

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