Eicosapentaenoic acid and docosahexaenoic acid

No. 2023/02De, The Hague, February 7, 2023

Background document to the advisory report:
Dutch dietary guidelines for people with atherosclerotic cardiovascular disease
No. 2023/02e, The Hague, February 7, 2023

Health Council of the Netherlands

Contents

1 Introduction

This background document belongs to the advisory report *Dutch dietary guidelines for people with atherosclerotic cardiovascular disease* (ASCVD).[1] It describes the methodology for the search, selection and evaluation of the literature regarding the relationship of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) with health outcomes in people with ASCVD. It also describes the scientific evidence on this topic and the conclusions that have been drawn by the council's Committee on Nutrition.

1.1 Definitions of eicosapentaenoic acid and docosahexaenoic acid

Eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are omega-3 (n-3) long-chain polyunsaturated fatty acids (PUFAs). EPA and DHA are also known as fish fatty acids, since fish is the most important source of EPA and DHA from the diet. Fatty fish contains more EPA and DHA than lean fish[a]: one portion of fatty fish (~100 grammes [g]) contains on average 2.5 g of EPA and DHA combined, and one portion of lean fish contains on average 0.4 g of EPA and DHA.[2] EPA and DHA are also added to foods, such as margarines. Fish oil supplements are also available, which mostly contain both EPA and DHA. In general, the dose of EPA and DHA in such supplements is much higher than the amount that can be obtained from the (Dutch) diet.

1.2 Dietary reference value for EPA/DHA and intake in the Netherlands

According to the Council's 2001 report on dietary reference values for energy and macronutrients, the adequate intake for n-3 fatty acids from fish for adults is 200 milligrammes per day (mg/d).[3]

For the *Dutch dietary guidelines 2015*, the Committee used the evidence on the relationship of EPA and DHA intake with health outcomes, together with the evidence on fish consumption, to formulate the dietary guideline on fish consumption: eat one serving of fish weekly, preferably oily fish.[4]

According to the Dutch National Food Consumption Survey 2012-2016, the median EPA and DHA intake of persons aged 1 to 79 years in the Netherlands is 107 mg/d (95%CI 101, 113). The mean intake is 158 mg/d (95%CI 149, 167).[5] Main dietary sources of EPA and DHA include fish (20%), meat (19%) and dairy products (16%). Fish oil supplements contribute little to the daily intake of EPA and DHA (5% on average).[5,6]

[a] Fatty fish is defined as fish that contains more than 5 g of fat per 100 g. According to the Dutch Food Composition Database, fatty fish contains 19 g of fat on average. Lean fish contains a maximum of 5 g of fat per 100 g (1.5 g on average).[2]

1.3 Safety

There is no report from the European Food Safety Authority (EFSA) or European Commission (EC) on the safety of general fish oil supplements. There is, however, an EFSA report on the tolerable upper intake level (in Dutch: *aanvaardbare bovengrens*) of n-3 PUFAs (EPA, DHA and docosapentaenoic acid [DPA]).[7] The main findings from this report are described below: EFSA reports that there is no evidence of adverse effects of n-3 PUFAs in amounts equal to what can be achieved with a usual diet (up to approximately 1 gramme per day [g/d]) in healthy adults. EFSA also sees no indications that long-term use of n-3 PUFAs (mostly EPA and DHA) with doses of up to 5 g/d increases the risk of adverse effects, such as bleeding, reduced immune function and reduced glucose metabolism. There are indications that a high dose of EPA and DHA (2 to 6 g/d of EPA and DHA combined and 2 to 4 g/d of DHA alone) can increase low-density lipoprotein (LDL) cholesterol levels by 3%. However, according to EFSA, this may not lead to an increased risk of cardiovascular disease (CVD). Supplementation with EPA alone in doses of up to 4 g/d likely has no effect on LDL cholesterol. Overall, EFSA concludes that available data are insufficient to establish a tolerable upper intake level for EPA and DHA, but also that, for the general population, there is no reason for concern about the safety of intakes of EPA and DHA supplements at doses of up to 5 g/d and of intakes of EPA supplements at doses of up to 1.8 g/d.

2 Methodology

2.1 Research questions

The Committee used the scientific evidence on EPA and DHA for two purposes. First, it used the evidence on associations or effects of EPA and DHA intake in amounts that can be achieved with a usual (Dutch) diet as ancillary evidence – secondary to the evidence on fish consumption – to evaluate the guideline on fish. In the Netherlands, fish is generally consumed once a week or less and at most a few times per week. Therefore, the Committee considers an EPA and DHA intake of no more than 1 g/d as the intake level that is feasible with the intake from fish. Second, the Committee used the evidence on relatively high amounts of EPA and DHA (>1 g/d) to evaluate whether supplementation with EPA and DHA should be advised to people with ASCVD. Therefore, the Committee specified two separate research questions:

1 What is the relationship (effect or association) of EPA and DHA intake up to and including 1 g/d with health outcomes in people with ASCVD?
2 What is the relationship (effect or association) of EPA and DHA intake of more than 1 g/d with health outcomes in people with ASCVD?

In contrast to the *Dutch dietary guidelines 2015*, where all evidence on EPA and DHA intake, regardless of the amount of EPA and DHA, was considered when formulating the guideline on fish, the Committee now considers that only the evidence on EPA and DHA intake in doses up to and including 1 g/d should be considered when evaluating the guideline on fish. As explained above, this intake level of EPA and DHA is feasible with the intake of fish (approximately 2-3 portions of fatty fish per week). Intake levels of 3 or 4 g/d EPA and DHA per day, for example, are far above the level that can be achieved with fish consumption (as part of a usual diet) and are generally achieved with supplements.

2.2 Target group

The target group of the current advisory report is people with ASCVD. The Committee defines this group as people with clinically established coronary heart disease (CHD, consisting of acute coronary syndromes [myocardial infarction and unstable angina], stable angina and revascularisation procedures such as percutaneous coronary intervention [PCI] and coronary artery bypass grafting [CABG]), peripheral arterial disease (PAD) or cerebrovascular disease (consisting of stroke and transient ischemic attack [TIA]). In the target population, atherosclerosis in the coronary arteries, aorta, iliac and femoral arteries, and cerebral arteries is the main underlying pathological process. Groups with a high risk (but no manifestation) of ASCVD, such as people with hypertension or elevated LDL cholesterol levels, fall outside this definition. Also, the target group of this advice does not include people with heart failure (except when

those people also suffer from ASCVD). A detailed description of the target group of this advisory report is provided in the background document *Methodology for the evaluation of the evidence*.[8]

In the present background document, the Committee also considered studies performed in people with CVD in general (not further specified), under the assumption that the majority of this population will have ASCVD.

2.3 Nutritional topics

The Committee searched for studies into supplementation with EPA and/or DHA. EPA and DHA are usually provided as a pill or capsule and are known as omega-3 supplements or fish oil supplements. The Committee included RCTs that used either a general fish oil supplement or a supplement of a highly purified form of EPA (icosapent ethyl [IPE]). In the European Union these products are usually classified as food supplements.[9] Certain compositions and dosages of omega-3 PUFAs can also be classified as a medicine. This is the case when the European Medicines Agency (EMA) and/or the Dutch Medicines Evaluation Board (in Dutch: *College ter Beoordeling van Geneesmiddelen*) has judged that it has been proven that a certain composition and dosage of omega-3 PUFAs has a specific therapeutical effect and that the risk-benefit balance of this specific product is favourable. In such a case, the specific product can be authorised to be placed on the market as medicinal product.[10-12] (For example, the EC has granted a marketing authorisation for the medicinal product Vazkepa (IPE), that (in a daily dose of four 1-g capsules) is indicated to reduce the risk of cardiovascular events in statin-treated adults at high cardiovascular risk, with elevated triglycerides and either established CVD or diabetes and at least one other cardiovascular risk factor. The EC's decision followed the positive opinion of the EMA, which was mainly based on one RCT with hard clinical endpoints in people with (a high risk of) CVD and two RCTs with surrogate endpoints in people with hypertriglyceridaemia.[12])
As omega-3 supplements are generally regarded as food supplements, the Committee included alle types of omega-3 supplements in its evaluation of EPA and DHA and evaluated the evidence using the methodology for foods (as described in the current document and the background document *Methodology for the evaluation of the evidence*[8]). The Committee notes that it may be slightly more questionable whether highly purified forms of EPA, such as IPE, can be regarded as food supplements. However, this involved the vast minority of evaluated studies. The Committee has therefore included RCTs investigating effects of IPE in its evaluation and evaluated whether there are indications that effects of IPE differ from effects of other types of EPA and DHA supplements.

Fish is the most important source of EPA and DHA, but it contains other nutrients and substances as well. The scientific evidence on fish consumption is described in the background document *Fish.*[13]

2.4 Health outcomes

The Committee selected the following health outcomes for this advisory report (further explained in the background document *Methodology for the evaluation of the evidence*[8]):

- short-term surrogate outcomes:
  - body weight
  - systolic blood pressure
  - low-density lipoprotein (LDL) cholesterol
  - estimated glomerular filtration rate (eGFR)
  - glycated haemoglobin (HbA1c) and fasting blood glucose
- long-term health outcomes:
  - all-cause mortality
  - morbidity and/or mortality from total CVD, CHD, stroke (cerebrovascular disease), heart failure, atrial fibrillation, type 2 diabetes, chronic obstructive pulmonary diseases (COPD), total cancer, breast cancer, colorectal cancer, lung cancer, dementia, depression
  - subtypes of CHD, such as MI, angina pectoris and revascularisation procedures (i.e. CABG and PCI)

In line with the approach taken for the *Dutch dietary guidelines 2015*, the Committee aimed to evaluate the evidence for fatal CHD, non-fatal CHD and sudden (cardiac) death separately, since there are indications that fish and fish fatty acids in particular protect against fatal CHD and sudden (cardiac) death.[14]

For cohort studies, the Committee included only studies in the above-described category named long-term health outcomes.

2.5 Selection and evaluation of the literature and drawing conclusions

2.5.1 Literature search

A detailed description of the approach used by the Committee for selecting and evaluating the scientific literature is provided in the background document *Methodology for the evaluation of the evidence*.[8] In short, the Committee aimed to base its evaluation of scientific literature on systematic reviews (SRs), including meta-analyses (MAs) and pooled analyses, of randomised controlled trials (RCTs) examining the effects of EPA and DHA supplementation on the above-mentioned health outcomes in people with ASCVD. To identify such publications, the Committee searched PubMed and Scopus in July 2021. In addition, the Committee searched for more recent individual RCTs that were not included in the most recent SR or MA. To this end,

PubMed and Scopus were searched in September 2021. The search strategies and specifications of the study selection are provided in Annex A.

2.5.2 Selection of randomised controlled trials

The Committee only found SRs and MAs of RCTs. For many health outcomes, multiple MAs were identified, which differed from each other with respect to included RCTs due to the time frame of the literature search and differences in eligibility criteria.

The Committee noted that most MAs included a large share of RCTs that did not fulfil the Committee's inclusion criteria. Because these RCTs constituted a large proportion of the pooled effect estimate, the Committee considered such MAs not suitable for its evaluation. The Committee also aimed to distinguish between the effects of EPA and DHA supplementation with 1 g/d or less and EPA and DHA supplementation with more than 1 g/d. In most MAs, this distinction was not made (usually all RCTs with different doses of EPA and DHA were lumped). Therefore, the Committee decided to describe each relevant RCT from the selected MAs separately. The RCTs from MAs were supplemented with additional individual RCTs (mostly more recent RCTs) that were not included in an MA. An overview of the RCTs that the Committee selected for its evaluation of the effect of EPA and DHA supplementation on health outcomes in people with ASCVD is given in Annex B.

The Committee notes that many of the conclusions in the *Dutch dietary guidelines 2015*[15] were based on studies performed in CVD patients. For some of the health outcomes evaluated in the current report, there is considerable overlap with the studies evaluated for the *Dutch dietary guidelines 2015* (but the evidence for the current report is supplemented by more recent RCTs and the primary prevention RCTs were disregarded).

2.5.3 Evaluation

Where it was possible and considered helpful to draw conclusions on the effects of EPA/DHA intake on health outcomes, the Committee pooled the results of all selected studies (i.e. the studies that met its inclusion criteria), using a random effects meta-analysis approach. Effect estimates of each individual study were obtained from the MAs. Only in cases where the effect estimate of an RCT could not be obtained from an MA was it obtained from the original publication. Because of the two research questions set for this dietary factor, the Committee reported the effects of EPA/DHA intake of ≤1 g/d (research question 1) separately from the effects of EPA/DHA intake of >1 g/d (research question 2).

Subgroup analyses according to EPA/DHA dose and statin use

To further examine whether any effects of EPA and DHA depend on the dose, the Committee performed subgroup analyses (at study level) among the RCTs with an EPA/DHA dose of >1 g/d, based on the cut-off of 3 g/d (corresponding to consumption of approximately one portion of fatty fish each day). This cut-off was predominantly based on the doses observed in the RCTs, so that sufficient studies remained in each category. Subgroup analyses according to dose were not performed among the RCTs with an EPA/DHA dose of ≤1 g/d, because too few RCTs were available to create subgroups.

The literature suggests that the effect of EPA/DHA may be less pronounced in people who are on statin therapy (to treat high LDL cholesterol levels) as compared to people who are not on statin therapy.[16,17] Therefore, where it was possible and considered appropriate (see below for more details), the Committee performed subgroup analyses according to statin use. Two subgroups were defined according to the proportion of participants in an RCT that used statins: <75% (low) and ≥75% (high). When information on the proportion of statin users was not available, the RCTs were classified according to the date (year) of execution of the trial. The Committee assumed that RCTs performed before 2000 included a low proportion of statin users and that RCTs performed since 2000 included a high proportion of statin users. These analyses were only performed among the RCTs with an EPA/DHA dose of >1 g/d, because too few RCTs with a dose of ≤1 g/d were available to create subgroups.

The Committee only performed subgroup analyses if there were at least 2 subgroups with at least 3 studies and 60 cases each (minimal requirements to draw a conclusion with limited evidence on hard clinical outcomes; for intermediate outcomes, at least 90 cases are required; see also section 2.5.5). If a strong indication for effect modification was found, the Committee drew separate conclusions for each subgroup. If not, the Committee drew one overall conclusion and described its observations regarding the subgroup evaluation in the explanatory text. The Committee used the ICEMAN (Instrument to assess the Credibility of Effect Modification Analyses) tool for MAs of RCTs as a tool to assist in assessing whether or not effect modification was present.[18] On the basis of eight items, the Committee assessed the likelihood that an apparent effect modification by EPA/DHA dose or statin use truly exists and that this is not the result of chance or bias. The items concern the following topics, amongst others: whether between- or within-study comparisons were made; whether the examination was based on an a priori hypothesis or was performed post-hoc (exploratory); the number of RCTs per subgroup; and whether cut points for subgroups (for this evaluation, this concerns cut-offs of 1 g/d and 3 g/d for EPA/DHA doses and a cut-off of 75% for the proportion of statin users) were chosen on an exploratory basis, based on previous research or based on a hypothesis.

EPA plus DHA versus EPA alone
Some suggest that only EPA, and not DHA or the combination of EPA and DHA, affects CVD risk.[19] The Committee has noted that most selected RCTs examined the effect of EPA plus DHA. Only four RCTs were found that examined the health effects of EPA alone.[20-23] Due to this limited number of RCTs, which moreover addressed only a few health outcomes, the Committee was only able to evaluate this hypothesis to a limited extent. For only one health outcome (total CVD), a sufficient number of RCTs was available to perform subgroup analyses according to intervention type (EPA alone versus EPA plus DHA).

Type of ASCVD
The Committee presents its findings and conclusions primarily for the total group of people with ASCVD (independent of the type of ASCVD) and describes whether results might vary according to subtypes of ASCVD. In the literature, the following subtypes of ASCVD are distinguished: people with CHD, people with stroke and people with peripheral artery disease (PAD).

Men versus women
The Committee aimed to evaluate whether effects of EPA and DHA supplements on health outcomes are similar in men and women.

2.5.4 Risk of bias assessment
For the majority of included studies, a risk of bias assessment was available in an existing MA. Where this was the case, the Committee did not systematically assess the risk of bias itself but used the risk of bias assessments that were presented in the MAs. For most RCTs, the Committee used the assessments from the MA by Abdelhamid et al., who described the risk of bias in detail.[24] The risk of bias of the RCTs for which no risk of bias assessment was available in an MA was assessed by the Committee using the revised Cochrane Collaboration's tool RoB2.[25]

The Committee noted that multiple MAs included the RCT by Singh et al.,[26] whereas it was excluded by some others. Reason for exclusion is that Singh is under suspicion of research misconduct[27] and the results of this RCT should therefore be interpreted with caution. The Committee did not exclude the RCT by Singh et al. in advance but judged and described for each analysis whether the RCT by Singh et al. might have substantially influenced the pooled result (similar to the way the risk of bias is considered in the assessment and interpretation of other studies).

2.5.5 Drawing conclusions
A detailed description of the approach used for drawing conclusions is provided in the background document *Methodology for the evaluation of the evidence*.[8] In short, the

Committee drew conclusions on (the certainty of) the evidence regarding the effects of EPA and DHA supplementation with risk of health outcomes in people with ASCVD, based on the number of studies, the number of participants and the number of cases that contributed to the evaluation. Also, it took the quality of the studies, in particular the risk of bias, and the heterogeneity between studies into account. The Committee used the decision tree (presented in the background document *Methodology for the evaluation of the evidence*[8]) as a tool to support consistency in drawing conclusions.

3 Effects of EPA and DHA

In this chapter, the Committee describes the scientific evidence for the effects of EPA and DHA supplementation on health outcomes in people with ASCVD. The scientific evidence is evaluated per health outcome and for EPA/DHA intake levels of ≤1 g/d and >1 g/d, separately. First, the results of the Committee's meta-analyses are presented in a table. Thereafter, the Committee's conclusions are presented, accompanied by a motivation based on the decision tree. The evaluation ends with a brief description and a quality assessment of the RCTs that the Committee selected for its evaluation. The characteristics and results of the selected RCTs are described in detail in Annex C.

3.1 Conclusions for EPA and DHA supplementation of ≤1 gramme per day

Here, the Committee describes the evaluation of the evidence from RCTs regarding the effects of supplementation of EPA and DHA with doses of ≤1 g/d in people with ASCVD. The results of the Committee's meta-analyses are presented in Table 1. In total, 6 RCTs with an EPA/DHA dose of ≤1 g/d, described in 8 publications, were found: Alpha Omega (2010),[28,29] GISSI-Prevenzione (GISSI-P, 2001),[30] Nutristroke (2009),[31] OMEGA (2010),[32] ORIGIN (2019)[33] and SU.FOL.OM3 (2010).[34,35]

Table 1 Results of the pooled analyses regarding effects of EPA and DHA supplementation with doses of ≤1 gramme per day on the risk of health outcomes in people with ASCVD: pooled RRs (95%CI) from RCTs, level of heterogeneity (I^2) and number of RCTs included (n)[a,b]

Health outcome	Main result
All-cause mortality	0.97 (0.83, 1.13), I^2=49%, n=5
Total CVD/MACE	0.99 (0.95, 1.04), I^2=0%, n=5
Fatal CVD	0.94 (0.77, 1.14), I^2=49%, n=5
Total CHD	0.94 (0.82, 1.08), I^2=17%, n=4
Fatal CHD	0.81 (0.70, 0.93), I^2=16%, n=4
Total MI	0.97 (0.85, 1.10), I^2=0%, n=4
Fatal MI	0.65 (0.36, 1.18), I^2=83%, n=4
Non-fatal MI	0.99 (0.87, 1.14), I^2=0%, n=4
Total stroke	1.20 (0.93, 1.54), I^2=24%, n=4
Sudden death	0.75 (0.61, 0.93), I^2=19%, n=3
Revascularisation	1.00 (0.92, 1.10), I^2=18%, n=3
Arrhythmia	1.01 (0.85, 1.21), I^2=0%, n=4
Cancer	1.17 (0.96, 1.44), I^2=0%, n=3

Abbreviations: ASCVD: atherosclerotic cardiovascular disease; CHD: coronary heart disease; CI: confidence interval, CVD: cardiovascular disease, DHA: docosahexaenoic acid, EPA: eicosapentaenoic acid, MACE: major adverse cardiovascular events, MI: myocardial infarction; n: number; RR: relative risk.

Footnotes:

[a] The table presents the results obtained from the meta-analyses of RCTs performed by the Committee.

[b] Outcomes for which the Committee did not perform a meta-analysis were not reported in the table.

3.1.1 All-cause mortality, total CVD and fatal CVD

The evaluation of the outcomes of all-cause mortality, total CVD and fatal CVD was based on the same five RCTs (Alpha Omega[28]; GISSI-P[30]; Nutristroke[31]; OMEGA[32]; SU.FOL.OM3[34]). In addition, the conclusions and motivation for these three outcomes are largely similar. For the sake of readability, the description of the evaluation of these three outcomes is therefore combined.

Based on the evaluation of RCTs, the Committee has concluded the following:
There is likely no effect of EPA and DHA supplementation of 0.4 to 0.88 grammes per day on the risks of all-cause mortality, total CVD and fatal CVD in people with ASCVD.

The following considerations were made by the Committee to come to these conclusions, following the steps of the decision tree:

1 Number of studies and cases:
There are 5 RCTs that addressed the effect of EPA/DHA supplementation with doses of ≤1 g/d on the risks of all-cause mortality, total CVD and fatal CVD in people with ASCVD.[28,30-32,34] For each outcome, more than 100 events were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of those 5 RCTs did not show an effect of EPA/DHA supplementation on the risks of all-cause mortality, total CVD or fatal CVD (Table 1). There is moderate heterogeneity between studies for the outcomes of all-cause mortality and fatal CVD (I^2=49% for both outcomes), which is most likely explained by the large GISSI-P trial. Only this RCT showed a statistically significant reducing effect on both all-cause mortality and fatal CVD. An explanation might be that only a few participants in the GISSI-P trial received statin therapy whereas nearly all participants did in the other four RCTs. There were, however, too few studies to perform subgroup analyses according to the proportion of statin users to further explore this hypothesis. No heterogeneity was observed between studies for the outcome of total CVD (I^2=0%).

3 Consideration regarding the quality of evidence:
The GISSI-P trial had an open-label design, but the Committee assumes that there is a low chance of performance bias (for an explanation, see section 3.3). Although the Committee expects any influence of performance bias to be small, it cannot rule out the possibility that some performance bias resulted in a slight

underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since mortality and CVD concern hard, clinical, non-subjective outcomes. The Nutristroke trial has a high risk of bias. However, this RCT made a very small contribution to the pooled analyses (<1%) and therefore likely did not substantially impact the overall findings. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
Three RCTs were performed in people with CHD,[28,30,32] one in people with stroke[31] and one in people with CHD or stroke.[34] The three RCTs in people with CHD tend to show, by approximation, a comparable result as compared to the RCTs in people with stroke (i.e. no effect). Therefore, and because there is no substantial heterogeneity between studies, the Committee sees no reason to expect differences in effect between people with CHD or stroke. Because studies in people with PAD were missing, the Committee could not evaluate whether the results observed also apply to this ASCVD subgroup.
In all RCTs, at least two-thirds of the study population comprised men. Results were not presented for men and women separately in those studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in those studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.1.2 Total CHD, total MI, fatal MI, non-fatal MI, total stroke and arrhythmia

The evaluation of the outcomes of total CHD, total MI, fatal MI, non-fatal MI, total stroke and arrhythmia was based on the same four RCTs (Alpha Omega[28]; GISSI-P[30]; OMEGA[32]; SU.FOL.OM3[34]). In addition, the conclusions and motivation for these outcomes are largely similar. For the sake of readability, the description of the evaluation of these five outcomes is therefore combined.

Based on the evaluation of RCTs, the Committee has concluded the following:
There is too little research to draw conclusions on the effect of EPA and DHA supplementation of 0.4 to 0.88 grammes per day on the risks of total CHD, total MI, fatal MI, non-fatal MI, total stroke and arrhythmia in people with ASCVD.

The following considerations were made by the Committee to come to these conclusions, following the steps of the decision tree:

1 Number of studies and cases:
There are 4 RCTs that addressed the effect of EPA and DHA supplementation (range: 0.4 to 0.88 g/d) on the risks of total CHD, total MI, fatal MI, non-fatal MI, total stroke and arrhythmia in people with ASCVD.[28,30,32,34] This excludes a

conclusion of ‘an effect is unlikely’ or a conclusion with strong evidence, for which at least 5 RCTs are required. In total, more than 100 cases of each outcome were reported.

2 Heterogeneity of the study findings:
Pooled analyses of those RCTs did not show an effect of EPA/DHA supplementation on the risks of total CHD, total MI, fatal MI, non-fatal MI, total stroke and arrhythmia in people with ASCVD (Table 1). No heterogeneity between studies (I^2=0%) was observed for the outcomes of total MI, non-fatal MI and arrhythmia, little heterogeneity (I^2<25%) for the outcomes of total CHD and total stroke and substantial heterogeneity (I^2=83%) for the fatal MI outcome.
The absence of obvious heterogeneity in the direction of the effects on total CHD, total MI, on-fatal MI, total stroke and arrhythmia in combination with the fact that the pooled estimate did not show an effect on these health outcomes and that four RCTs is too few to base the conclusion ‘likely no effect’ on, let the Committee downgrade its conclusion to ‘too little research’ for those five health outcomes.
For fatal MI, relative risks (RRs) of the 4 included RCTs varied between 0.36 and 1.16, 2 of the 4 RCTs showed a statistically significant reducing effect (RRs 0.36 and 0.80) and the other 2 RCTs showed no effect of EPA/DHA supplementation on fatal MI. The lack of significance in one of the RCTs (SU.FOL.OM3 trial[34]) might be due to the relatively small sample size and few fatal MI cases reported (3 in total). Besides sample size, another explanation for the heterogeneity could not be found. The Committee considered it unlikely that statin use or whether the participants had CHD or stroke at baseline explained the heterogeneity. Four RCTs is too few to draw a conclusion of ‘inconclusive evidence’, for which at least 5 studies are required. Therefore, and because there was no obvious heterogeneity in direction of the effects (according to the decision tree, heterogeneity in direction of the effects might argue for the conclusion 'contradictory evidence'), the Committee downgraded its conclusions and concluded that there is too little research.

3.1.3 Fatal CHD

Based on the evaluation of RCTs, the Committee has concluded the following:
EPA and DHA supplementation of 0.4 to 0.88 grammes per day reduces the risk of fatal CHD in people with ASCVD. The evidence is limited.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 4 RCTs that addressed the effect of EPA and DHA supplementation (range: 0.4 to 0.88 g/d) on the risk of fatal CHD in people with ASCVD (Alpha Omega[28]; GISSI-P[30]; OMEGA[32]; SU.FOL.OM3[34]). This excludes a conclusion of

'an effect is unlikely' or a conclusion with strong evidence, for which at least 5 RCTs are required. However, since in total more than 60 cases of fatal CHD were reported, a conclusion with limited evidence is still possible.

2 Heterogeneity of the study findings:
A pooled analysis of those RCTs showed that EPA/DHA supplementation reduced the risk of fatal CHD with 19% on average in people with ASCVD (Table 1). Little heterogeneity between studies was observed (I^2=16%). The Committee noted that the pooled result was largely driven by the GISSI-P trial (73%). Since the effect estimates of the other 3 RCTs point in the same direction (RR < 1.0), the Committee assumes that the pooled result provides sufficient evidence to base a conclusion with limited evidence on.

3 Consideration regarding the quality of evidence:
The GISSI-P trial had an open-label design, but the Committee assumes that there is a low chance of performance bias (for an explanation, see section 3.3). Although the Committee expects any influence of performance bias to be small, it cannot rule out the possibility that some performance bias resulted in a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since CHD concerns a hard, clinical, non-subjective outcome. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
Three RCTs were performed in people with CHD[28,30,32] and one in people with CHD or stroke.[34] Since Galan et al. (SU.FOL.OM3 trial[34]) noted that their trial might have been underpowered, it is difficult to compare the effect estimate of this RCT with those of the other RCTs. However, all RCTs in people with CHD tend to show, by approximation, a comparable result as compared to the RCT in people with CHD or stroke, i.e. a (tendency towards a) reducing effect. Therefore, the Committee sees no reason to expect differences in effect between people with CHD and people with stroke. Because studies in people with PAD were missing, the Committee could not evaluate whether the results observed also apply to this ASCVD subgroup.
In all RCTs, at least 74% of the study population comprised men. Results were not presented for men and women separately in those studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in those studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.1.4 Non-fatal CVD, angina pectoris, heart failure, PAD progression, depression, systolic blood pressure and LDL cholesterol

The conclusions and accompanying explanation for the long-term health outcomes of non-fatal CVD, angina pectoris, heart failure, PAD progression and depression and for the surrogate endpoints of systolic blood pressure and LDL cholesterol were largely similar. For the sake of readability, the description of the evaluation of these seven health outcomes is therefore combined.

Based on the evaluation of RCTs, the Committee has concluded the following:

- **There is too little research to draw conclusions regarding the effect of EPA and DHA supplementation of ≤1 gramme per day on the risks of non-fatal CVD, angina pectoris, heart failure, PAD progression and depression in people with ASCVD.**
- **There is too little research to draw conclusions regarding the effect of EPA and DHA supplementation of ≤1 gramme per day on systolic blood pressure and LDL cholesterol in people with ASCVD.**

The following considerations were made by the Committee to come to these conclusions, following the steps of the decision tree:

1 Number of studies and cases:
There is one RCT that addressed the effect of EPA/DHA supplementation of <1 g/d on the risk of non-fatal CVD in people with ASCVD (GISSI-P[36]). This RCT showed no effect of EPA and DHA supplementation of 0.88 g/d on the risk of non-fatal CVD in people with CHD. Two RCTs addressed the effect on the risk of angina pectoris (GISSI-P[30,36]; OMEGA[32]), both showing no effect of EPA and DHA supplementation of 0.85-0.88 g/d in people with CHD. Two RCTs addressed the risk of heart failure (SU.FOL.OM3[34]; OMEGA[32]), both showing no effect of EPA and DHA supplementation of 0.6-0.85 g/d in people with CHD or stroke. One RCT addressed the risk of EPA and DHA supplementation on PAD progression in people with PAD (ORIGIN[33]). No effect of 0.84 g/d of EPA and DHA was observed. Two RCTs addressed the risk of depression (Alpha Omega[28]; SU.FOL.OM3[34]) and showed partially conflicting results. One RCT showed no effect of EPA and DHA supplementation of 0.4 g/d in people with CHD,[29] whereas the other RCT showed that EPA and DHA supplementation of 0.6 g/d adversely affected depressive symptoms at follow-up in men, but not in women.[35] In the two RCTs addressing systolic blood pressure (Alpha Omega[28]; SU.FOL.OM3[34]), no effects of EPA and DHA supplementation in CHD or stroke patients were observed. Also, the single RCT examining the effect of EPA and DHA supplementation on LDL cholesterol in people with CHD, showed no statistically significant effect (Alpha Omega[28]). Less than 3 studies provide too little evidence to draw conclusions. Therefore, the Committee concluded that there was too little

research regarding the effects of EPA and DHA supplementation on the health outcomes of non-fatal CVD, angina pectoris, heart failure, depression, systolic blood pressure and LDL cholesterol.

3.1.5 Sudden death

Based on the evaluation of RCTs, the Committee has concluded the following:
EPA and DHA supplementation of 0.6 to 0.88 grammes per day reduces the risk of sudden death in people with ASCVD. The evidence is limited.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 3 RCTs that addressed the effect of EPA and DHA supplementation (0.6 to 0.88 g/d) on the risk of sudden death in people with ASCVD (GISSI-P[30]; OMEGA[32]; SU.FOL.OM3[34]). This excludes a conclusion of 'an effect is unlikely' or a conclusion with strong evidence, for which at least 5 RCTs are required. However, since in total more than 60 cases of sudden death were reported, a conclusion with limited evidence is still possible.

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs showed that EPA and DHA supplementation reduced the risk of sudden death with 25% (RR 0.75, 95%CI 0.61, 0.93). Little heterogeneity between studies was observed (I^2=19%; Table 1). The pooled effect was largely driven by the GISSI-P trial (80%). Since the effect estimates of the other 2 RCTs point in the same direction (RR < 1.0), the Committee assumes that the pooled result provides sufficient evidence to base a conclusion with 'limited evidence' on.

3 Considerations regarding the quality of the evidence:
The GISSI-P trial had an open-label design, but the Committee assumes that there is a low chance of performance bias (for an explanation, see section 3.3). Although the Committee expects any influence of performance bias to be small, it cannot rule out the possibility that some performance bias resulted in a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since sudden death concerns a hard, clinical outcome. The Committee noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
Two RCTs were performed in people with CHD[30,32] and the other in people with CHD or stroke.[34] Since Galan et al. (SU.FOL.OM3[34]) noted that their trial might have been underpowered, it is difficult to compare the effect estimate of this RCT with those of the other RCTs. However, this RCT in people with CHD or stroke tend to show, by approximation, a comparable result as compared to the RCTs in

people with CHD. Therefore, the Committee sees no reason to expect differences in effect between people with CHD and people with stroke. Because studies in people with PAD were missing, the Committee could not evaluate whether the results observed also apply to this ASCVD subgroup.
In all RCTs, at least 74% of the study population comprised men. Results were not presented for men and women separately in those studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in those studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.1.6 Revascularisation and cancer

The evaluation of the outcomes of revascularisation and cancer were based on the same three RCTs (GISSI-P[30,36]; OMEGA[32]; SU.FOL.OM3[34]). In addition, the conclusions and motivation for these outcomes were largely similar. For the sake of readability, the description of the evaluation of these two outcomes is therefore combined.

Based on the evaluation of RCTs, the Committee has concluded the following:
There is too little research to draw conclusions on the effect of EPA and DHA supplementation of ≤1 gramme per day on the risks of revascularisation and cancer in people with ASCVD.

The following considerations were made by the Committee to come to these conclusions, following the steps of the decision tree:

1 Number of studies and cases:
There are 3 RCTs that addressed the effect of EPA and DHA supplementation (range: 0.6 to 0.88 g/d) on the risks of revascularisation (mainly coronary revascularisation) and cancer in people with ASCVD (GISSI-P[30]; OMEGA[32]; SU.FOL.OM3[34]). This excludes a conclusion of 'an effect is unlikely' or a conclusion with strong evidence, for which at least 5 RCTs are required. However, since in total more than 60 cases per health outcome were reported, a conclusion with limited evidence is still possible.

2 Heterogeneity of the study findings:
Pooled analyses of those 3 RCTs did not show an effect of EPA and DHA supplementation on the risks of revascularisation and cancer (Table 1). Little heterogeneity between studies was observed for the outcome of revascularisation (I^2=18%) and no heterogeneity was observed for the outcome of cancer.
Three RCTs is too few to draw a conclusion of 'likely no effect'. Therefore, the Committee downgraded its conclusion and concluded (based on the decision tree) that there is too little research.

3.1.7 General finding regarding the RCTs with an EPA/DHA dose of ≤1 gramme per day

The Committee noted that only one of the RCTs that examined the effect of a relatively low dose of EPA and DHA (≤1 g/d) included a subgroup analysis that was exclusively performed in people who do not or only occasionally consume fish. This concerned the Alpha Omega trial.[28] This RCT showed that effects of EPA and DHA supplementation on total CVD were not different in people with a low background intake of fish (<5 g/d) compared to those with a relatively high background fish intake (≥5 g/d). Given the scarcity of data, the Committee judged that it could not properly evaluate whether or not a supplement with a low dose of EPA and DHA (an amount comparable to 1 or 2 portions of fish) could be recommended for people with ASCVD who do not eat fish.

3.2 Conclusions for EPA and DHA supplementation of >1 gramme per day

Here, the Committee describes the evaluation of the evidence from RCTs regarding the effects of supplementation of EPA and DHA with doses of >1 g/d in people with ASCVD. The results of the Committee's meta-analyses are presented in Table 2. In total, 17 RCTs with an EPA and DHA dose of >1 g/d, described in 19 publications, were found: Gans et al. (1990),[37] HARP (1995),[38] HEARTS (2017),[39] IEIS-4 (1997),[26] JELIS (2007),[23,40] Mori et al. (1992),[41] NAT2 (2013),[42] Nosaka et al. (2017),[21] Nye et al. (1990),[22] OFAMI (2001),[43] OMEGA-REMODEL (2016),[44] OMEMI (2021),[45] OPACH (2006),[46] REDUCE-IT (2019),[20,47] SCIMO (1999),[48] SHOT (1996)[49] and STRENGTH (2020).[50]

Table 2 Results of the pooled analyses regarding effects of EPA and DHA supplementation with doses of >1 gramme per day on the risk of health outcomes in people with ASCVD: pooled RRs (95%CI) from RCTs, level of heterogeneity (I^2) and number of RCTs included (n)[a,b,c]

Health outcome	Main result	Results according to EPA/DHA dose[d]	Results according to proportion of statin users[e]	Results according to intervention type[f]
All-cause mortality	0.92 (0.69, 1.22), I^2=41%, n=12	DOSE >1 TO <3 G/D: 0.72 (0.45, 1.14), I^2=48%, n=6 DOSE ≥3 G/D: 1.18 (0.98, 1.41), I^2=0%, n=6	LOW: 0.86 (0.57, 1.29), I^2=26%, n=6 HIGH: 1.04 (0.77, 1.42), I^2=50%, n=6	N/A
Total CVD/MACE	0.89 (0.78, 1.01), I^2=61%, n=12	DOSE >1 TO <3 G/D: 0.83 (0.68, 1.02), I^2=56%, n=6 DOSE ≥3 G/D: 0.94 (0.77, 1.15), I^2=70%, n=6	LOW: 0.94 (0.76, 1.16), I^2=17%, n=6 HIGH: 0.86 (0.73, 1.02), I^2=75%, n=6	EPA PLUS DHA: 0.97 (0.89, 1.06), I^2=0%, n=9 EPA ALONE: 0.73 (0.67, 0.81), I^2=17%, n=3
Fatal CVD	1.09 (0.88, 1.35), I^2=0%, n=6	N/A	N/A	N/A

Non-fatal CVD	0.67 (0.27, 1.64), I^2=75%, n=3	N/A	N/A	N/A
Total CHD	0.85 (0.77, 0.94), I^2=36%, n=9	DOSE >1 TO <3 G/D: 0.67 (0.50, 0.92), I^2=12%, n=5 DOSE ≥3 G/D: 0.95 (0.76, 1.19), I^2=45%, n=4	LOW: 0.66 (0.42, 1.02), I^2=48%, n=6 HIGH: 0.86 (0.77, 0.95), I^2=25%, n=3	N/A
Fatal CHD	0.76 (0.51, 1.14), I^2=0%, n=6	N/A	N/A	N/A
Total MI	0.69 (0.42, 1.15), I^2=34%, n=7	N/A	N/A	N/A
Fatal MI	0.73 (0.43, 1.24), I^2=0%, n=5	N/A	N/A	N/A
Non-fatal MI	0.88 (0.60, 1.28), I^2=28%, n=8	N/A	LOW: 0.83 (0.44, 1.59), I^2=43%, n=5 HIGH: 0.93 (0.58, 1.47), I^2=20%, n=3	N/A
Total stroke	1.09 (0.63, 1.90), I^2=47%, n=8	N/A	LOW: 2.11 (0.78, 5.70), I^2=26%, n=4 HIGH: 0.85 (0.59, 1.22), I^2=31%, n=4	N/A
Sudden death	0.72 (0.32, 1.65), I^2=23%, n=5	N/A	N/A	N/A
Revasculari-sation	0.73 (0.61, 0.87), I^2=0%, n=8	DOSE >1 TO <3 G/D: 0.82 (0.66, 1.01), I^2=0%, n=5 DOSE ≥3 G/D: 0.66 (0.55, 0.79), I^2=0%, n=3	LOW: 0.84 (0.58, 1.22), I^2=0%, n=4 HIGH: 0.70 (0.56, 0.88), I^2=36%, n=4	N/A
Angina pectoris	0.63 (0.41, 0.98), I^2=56%, n=6	N/A	N/A	N/A
Heart failure	1.06 (0.59, 1.90), I^2=0%, n=3	N/A	N/A	N/A
Systolic blood pressure	MD (in mmHg): -0.48 (-2.32, 1.37), I^2=0%, n=6	N/A	N/A	N/A
LDL cholesterol	MD (in mmol/L): 0.06 (-0.01, 0.14), I^2=49%, n=8	N/A	MD (in mmol/L): LOW: 0.29 (0.04, 0.55), I^2=54%, n=5	N/A

			HIGH: 0.02 (-0.05, 0.10), I^2=0%, n=3	
Body weight	MD (in kg): 0.15 (-1.25, 1.56), I^2=0%, n=3	N/A	N/A	N/A

Abbreviations: ASCVD: atherosclerotic cardiovascular disease; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; LDL: low-density lipoprotein; MACE: major adverse cardiovascular events; MD: mean difference; N/A: not applicable; RCT: randomised controlled trial; RR: relative risk.

Footnotes:

[a] The table presents the results obtained from the meta-analyses of RCTs performed by the Committee.

[b] Outcomes for which the Committee did not perform a meta-analysis were not reported in the table.

[c] For the outcomes of systolic blood pressure, LDL cholesterol and body weight, the results are presented as between-group MDs instead of RRs.

[d] Results are presented for the subgroup of RCTs with an EPA/DHA dose of >1 to <3 g/d and for the subgroup of RCTs with an EPA/DHA dose of ≥3 g/d, where applicable.

[e] Results are presented for the subgroup of RCTs of which <75% of the study population used statins (indicated as 'low') and for the subgroup of RCTs of which ≥75% of the study population used statins (indicated as 'high'), where applicable.

[f] Results are presented for the subgroup of RCTs that examined the effect of EPA and DHA combined (indicated as 'EPA plus DHA') and for the subgroup of RCTs that examined the effect of EPA alone, where applicable.

3.2.1 All-cause mortality

Based on the evaluation of RCTs, the Committee has concluded the following:

There is likely no effect of EPA and DHA supplementation of 1.8 to 4.8 grammes per day on the risk of all-cause mortality in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 12 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.8 to 4.8 g/d) on the risk of all-cause mortality in people with ASCVD.[21,26,38,39,42-46,48-50] In total, more than 100 cases were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of those 12 RCTs did not show an effect of EPA and DHA supplementation on the risk of all-cause mortality (Table 2). There is moderate heterogeneity between studies (I^2=41%). Moderate heterogeneity was observed in the direction of the effect, which seems to be predominantly caused by three RCTs: two RCTs (IEIS-4 trial[26] and Nosaka et al.[21]) showed a strong statistically significant reducing effect, whereas one other RCT (STRENGTH trial[50]) showed a borderline significant increasing effect. Differences between these studies were

observed with regard to sex, patient type and/or EPA and DHA dose, which may contribute to the heterogeneity. The IEIS-4 trial and the RCT by Nosaka et al. were conducted in men alone, whereas 35% of the participants of the STRENGTH trial were female. The STRENGTH trial was conducted in people with all types of CVD, whereas the other 2 RCTs were conducted in people with CHD. Lastly, the dose of EPA and DHA was much larger in the STRENGTH trial (4 g/d) compared to the other RCTs (both 1.8 g/d).

Subgroup analyses according to EPA and DHA dose showed no effect of EPA and DHA supplementation of >1 to <3 g/d on the risk of all-cause mortality, which is similar to the main result. Moderate to substantial heterogeneity between studies remained within this subgroup (consisting of 6 RCTs). For EPA and DHA supplementation of ≥3 g/d, a trend towards an increased mortality risk was observed (RR 1.18, 95%CI 0.98, 1.41). This result was largely (90%) driven by one RCT (STRENGTH trial), which may also explain why the I^2 was 0% while the forest plot shows heterogeneity in the size of the effect between the 6 RCTs within this subgroup. Considering the above observations and by using the ICEMAN tool for assessing the credibility of effect modification, the Committee considered that there is no strong indication for effect modification by EPA/DHA dose, for the following main reasons: 1) the result for EPA and DHA supplementation of ≥3 g/d is predominantly based on only 1 RCT; 2) there is no clear hypothesis as to why relatively higher intake levels of EPA/DHA would increase the risk of all-cause mortality; 3) the level of heterogeneity did not reduce within the subgroup of RCTs with a dose of >1 to <3 g/d; and 4) the cut-off of 3 g/d was pragmatically chosen based on the doses used in the available RCTs and not based on a pre-defined hypothesis.

Subgroup analyses according to the proportion of statin users did not show an effect among the 6 RCTs with a low proportion of statin users. Moderate heterogeneity between studies was observed in this subgroup (I^2=26%). The 6 RCTs with a relatively high proportion of statin users showed no effect of EPA and DHA supplementation on mortality risk either, but substantial heterogeneity (I^2=50%) remained between the studies within this subgroup. The heterogeneity might be partially due to the relatively large contribution of the STRENGTH trial (61%) that tend to show an increasing effect while the other RCTs showed neutral or reducing effects. Based on the above results and by using the ICEMAN tool, the Committee considered that there is no strong indication for effect modification by statin use, for the following main reasons: 1) the observed effect (no effect) was similar in both subgroups; and 2) the result for the subgroup of RCTs with a high proportion of statin users is largely based on only 1 RCT.

To conclude, the Committee could not specify one factor that fully explains the heterogeneity observed, and assumes that the heterogeneity is most likely due to multiple differences between studies.

3 Consideration regarding the quality of evidence:
There are some considerations regarding the validity of the results of the RCT by Singh,[26] who was suspected of research misconduct. However, the Committee expects that exclusion of this RCT will likely not substantially affect the pooled estimate. Therefore, the Committee did not exclude this RCT. For two RCTs with an open-label design (HEARTS[39] and Nosaka et al.[21]) it could not be judged to what extent this may have introduced performance bias. However, those RCTs together contributed less than 5% to the MA, and the Committee therefore expects they did not substantially impact the results. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
Almost all RCTs (n=9) were performed in people with CHD, and three RCTs[42,46,50] were performed in people with total CVD. The three RCTs in people with CVD (not further specified) tend to show, by approximation, a comparable result as compared to the RCTs in people with CHD (i.e. no effect). Therefore, and because there is no substantial heterogeneity between studies, the Committee sees no reason to expect differences in effect between people with different types of ASCVD.
In almost all RCTs, at least two-thirds of the study population comprised men. Results were not presented for men and women separately in those studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in those studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.2.2 Total CVD

Based on the evaluation of RCTs, the Committee has concluded the following:

- **There is likely no effect of EPA and DHA supplementation of 1.1 to 4.8 grammes per day on the risk of total CVD in people with ASCVD.**
- **EPA supplementation of 1.8 to 4.0 grammes per day reduces the risk of total CVD in people with ASCVD. The evidence is limited.**

The following considerations were made by the Committee to come to these conclusions, following the steps of the decision tree:

1 Number of studies and cases:
There are 12 RCTs that addressed the effect of EPA/DHA supplementation on the risk of total CVD in people with ASCVD.[20,21,23,26,38,39,43,45,46,48-50] In total, more than 100 events were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of those 12 RCTs showed a borderline significant reducing effect of EPA and DHA supplementation on the risk of total CVD (Table 2). Substantial heterogeneity between studies was observed (I^2=61%). Three RCTs (JELIS,[23] REDUCE-IT[20] and Nosaka et al.[21]) showed protective effects and the other RCTs did not show an effect. The Committee assumes that the most likely explanation for the heterogeneity is the type of the intervention (EPA plus DHA versus EPA alone) since the three RCTs that show an effect examined the effect of EPA alone while the other RCTs examined the effect of EPA plus DHA. The pooled analysis of the three RCTs that examined the effect of supplementation of EPA alone, showed that this reduced the risk of CVD by an average of 27%. Heterogeneity between these studies (I^2) was reduced to 17%. The pooled analysis of the nine RCTs that examined the effect of supplementation of EPA plus DHA did not show an effect on CVD risk and no heterogeneity remained between these studies (I^2=0%). Based on the above observations and by using the ICEMAN tool, the Committee considered that there is a moderate indication for effect modification by intervention type, for the following main reasons: 1) both subgroups included at least three RCTs (several of which are large RCTs) and there is little heterogeneity between studies within this subgroup; and 2) examining the presence of effect modification by intervention type was based on an a priori hypothesis (previous studies suggested that EPA, and not DHA, might be responsible for the cardioprotective effect[19,51]) and the result is in line with that hypothesis. Because of this presumed effect modification by intervention type, the Committee drew two separate conclusions, one for the effect of EPA alone and one for the effect of EPA plus DHA. As there are only three RCTs examining the effect of EPA alone, no conclusion with strong evidence could be drawn for this exposure.

3 Considerations regarding the quality of the evidence:
There are some considerations regarding the validity of the results of the RCT by Singh, who was suspected of research misconduct. However, the contribution of the RCT by Singh et al. to the pooled result for EPA plus DHA was relatively small (<5%) and the result of this RCT was by approximation in line with the pooled result. Therefore, the Committee expects that this RCT did not substantially affect the pooled estimate. Four RCTs had an open-label design. This concerns 2 RCTs that examined the effect of EPA alone (JELIS[23] and Nosaka et al.[21]) and 2 RCTs that examined the effect of EPA plus DHA (HEARTS[39] and SHOT[49]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in

a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since CVD concerns a hard, clinical, non-subjective outcome. Overall, the Committee assumes that there is a low chance that the open-label design (and not the type of intervention) fully explains the reducing effect among the RCTs examining EPA alone. The HEARTS and SHOT trials contributed relatively little (6.8%) to the pooled result for EPA plus DHA, and the Committee therefore expects they did not substantially impact the results. Lastly, in the OMEMI trial, all-cause mortality was included in the definition of MACE, so the results of this RCT might be partially explained by non-CVD-related events (also given the fact that participants were 70-80 years old at baseline). Whether or not these deaths were CVD-related is unknown and it is thus unclear how this might have affected the effect observed. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
From the nine RCTs that examined the effect of EPA plus DHA, seven RCTs[26,38,39,43,45,48,49] were performed in people with CHD and two RCTs[46,50] in people with CVD. The two RCTs in people with CVD (not further specified) tend to show, by approximation, a comparable result as compared to the RCTs in people with CHD (i.e. no effect). Therefore, and because there is no heterogeneity between studies observed, the Committee sees no reason to expect differences in effects of EPA and DHA supplementation between people with different types of ASCVD. From the three RCTs that examined the effect of EPA alone, two RCTs[21,23] were performed in people with CHD and one RCT[20] was performed in people with CVD. Because the results of these RCTs were comparable and little heterogeneity between these RCTs was observed, the Committee sees no reason to expect differences in effects of EPA supplementation between people with different types of ASCVD.
In all twelve RCTs evaluated for this health outcome, at least two-thirds of the study population comprised men. Results were not presented for men and women separately in these studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in these studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.2.3 Fatal CVD

Based on the evaluation of RCTs, the Committee has concluded the following: **There is likely no effect of EPA and DHA supplementation of 1.8 to 4.8 grammes per day on the risk of fatal CVD in people with ASCVD.**

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 6 RCTs that addressed the effect of EPA and DHA supplementation (range: 1.8 to 4.8 g/d) on the risk of fatal CVD in people with ASCVD. In total, more than 100 cases were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of those RCTs did not show an effect of EPA and DHA supplementation on the risk of fatal CVD (Table 2). There was no heterogeneity between studies (I^2=0%). The Committee noted that the pooled result was largely driven by the STRENGTH trial (89%). Since the other five RCTs tend to show comparable results (i.e. no effect) and given the absence of heterogeneity, the Committee assumes that the pooled result provides sufficient evidence to base a conclusion on.

3 Considerations regarding the quality of the evidence:
For two RCTs with an open-label design it could not be judged to what extent this may have introduced performance bias (SHOT and Nosaka et al.). However, those RCTs together contributed little to the pooled result (4.7%), and therefore the Committee expects it did not substantially impact the results. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
Almost all RCTs (n=5) were performed in people with CHD. Only the STRENGTH trial, the RCT that made a significant contribution to the pooled result, was performed in people with (any type of) CVD. All RCTs show, by approximation, a comparable result. Therefore, and because there is no heterogeneity between studies, the Committee sees no reason to expect differences in effect between people with different types of ASCVD.
In almost all RCTs at least two-thirds of the study population comprised men. Results were not presented for men and women separately in these studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in these studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.2.4 Non-fatal CVD

Based on the evaluation of RCTs, the Committee has concluded the following:
There is too little research to draw conclusions on the effect of EPA and DHA supplementation of >1 grammes per day on the risk of non-fatal CVD in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 3 RCTs that addressed the effect of EPA and DHA supplementation (range: 1.8 to 3.5 g/d) on the risk of fatal CVD in people with ASCVD.[26,43,48] This excludes a conclusion of 'an effect is unlikely' or a conclusion with strong evidence, for which at least 5 RCTs are required. However, since 3 RCTs with in total more than 60 cases were available, a conclusion with limited evidence is still possible.

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA and DHA supplementation on the risk of non-fatal CVD (Table 2). There was substantial heterogeneity between studies (I^2=75%). One RCT[26] showed that EPA and DHA supplementation substantially and statistically significantly reduced the risk of non-fatal CVD by 56%, whereas the other RCTs[43,48] did not show an effect. Three RCTs is too few to draw a conclusion of 'likely no effect' or 'inconclusive evidence'. Therefore, and because there was no obvious heterogeneity in direction of the effect, the Committee downgraded its conclusions and concluded (based on the decision tree) that there is too little research.

3.2.5 Total CHD

Based on the evaluation of RCTs, the Committee has concluded the following:
EPA and DHA supplementation of 1.4 to 4 grammes per day reduces the risk of total CHD in people with ASCVD. The evidence is strong.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 9 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.4 to 4 g/d) on the risk of total CHD in people with ASCVD.[22,23,26,39,43,46,48-50] In total, more than 100 cases were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs showed that EPA/DHA supplementation reduced

the risk of total CHD by an average of 15% (Table 2). There was moderate heterogeneity between studies (I^2=36%). Both neutral and protective effects were observed. Three RCTs[23,46,50] showed a (borderline) reducing effect and the other six RCTs[22,26,39,43,48,49] did not show an effect of EPA/DHA supplementation on total CHD.

The Committee noted that the studies with EPA/DHA doses of 1-3 g/d generally showed a stronger reduction in total CHD risk than those with doses of ≥3 g/d (Table 2). However, moderate heterogeneity remained in the subgroup of studies with EPA/DHA doses of ≥3 g/d (I^2=45%). Furthermore, the Committee noted that there is no clear hypothesis for a U-shaped effect of EPA/DHA on the risk of total CHD (NB: no effect on total CHD was observed for an EPA/DHA dose of ≤1 g/d; Table 1). Also, the cut-offs of 1 and 3 g/d were predominantly pragmatically chosen based on the doses used in the available RCTs and not based on a pre-defined hypothesis. Based on these observations and by using the ICEMAN tool for assessing the credibility of effect modification, the Committee considered that there is no strong indication for effect modification by EPA/DHA dose. Thus, the Committee assumes that the heterogeneity is not fully explained by the EPA/DHA dose administered.

The Committee also assumes that the proportion of statin users did likely not fully explain the heterogeneity since in subgroup analyses according to proportion of statin users, moderate heterogeneity remained in both subgroups (I^2=25-48%). Furthermore, the Committee saw no clear indication that the type of intervention (EPA alone versus EPA plus DHA) explains the heterogeneity. However, since only two RCTs examined the effect of EPA only,[22,23] the Committee cannot rule out the possibility that EPA alone has a different effect than EPA and DHA combined. To conclude, the Committee could not specify one factor that fully explains the heterogeneity observed and assumes that the moderate heterogeneity is most likely due to multiple differences between studies.

3 Considerations regarding the quality of the evidence:

There are some considerations regarding the validity of the results of Singh, who was suspected of research misconduct. However, the RCT by Singh et al. contributed only 3% to the pooled analysis and would therefore not substantially impact the pooled estimate. For three RCTs with an open-label design it could not be judged to what extent this may have introduced bias (HEARTS,[39] JELIS[23] and SHOT[49]). In addition, the authors of the JELIS trial noted that their trial was likely underpowered, suggesting that the borderline effect observed might be an underestimation of the true effect. The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some

performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since CHD concerns a hard, clinical, non-subjective outcome. The Committee has noted no other major considerations regarding the quality of the RCTs that may impact the overall findings. According to the decision tree, in the case of no substantial heterogeneity and in the absence of major considerations, the evidence is judged as strong. Because of the moderate heterogeneity observed in the size of the effect, the Committee decided, however, not to quantify the effect in its conclusion.

4 Generalisability:

Seven RCTs included people with CHD and two RCTs[46,50] included people with (any type of) CVD. The Committee considered that the reducing effects tend to be more pronounced in the RCTs performed in people with any type of CVD than among the RCTs performed in people with CHD. This may suggest that EPA and DHA supplementation may be more beneficial in people with a type of CVD other than CHD. However, in the large STRENGTH trial[50] approximately 80% of the participants with established CVD had CHD (and 15% had cerebrovascular disease and 5% PAD). This trial showed a statistically significant reducing effect, which indicates that EPA and DHA supplementation is likely also effective in CHD patients. Based on this finding, in combination with the lack of substantial heterogeneity between the studies included in this pooled analysis, the Committee assumes that the effects of EPA and DHA supplementation on total CHD do not substantially differ between people with various types of ASCVD.

The Committee has noted that in almost all RCTs at least two-thirds of the study population comprised men. Whether results differed between men and women was not assessed in those studies and results were not presented for men and women separately in those studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in those studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.2.6 Fatal CHD

Based on the evaluation of RCTs, the Committee has concluded the following:

There is likely no effect of EPA and DHA supplementation of 1.8 to 4.8 grammes per day on the risk of fatal CHD in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 6 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.8 to 4.8 g/d) on the risk of fatal CHD in people with ASCVD.[23,26,38,43,48,49] In total, more than 100 cases were reported (n=108). This is the first step required to mark the evidence as strong or to allow conclusions of 'an effect is unlikely' or 'inconclusive evidence' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these 6 RCTs did not show an effect of EPA and DHA supplementation on the risk of fatal CHD (Table 2) and no heterogeneity between studies was observed (I^2=0%).

3 Consideration regarding the quality of evidence:
There are some considerations regarding the validity of the results of the RCT by Singh, who was suspected of research misconduct. However, the Committee expects that exclusion of this RCT (the only one showing an effect) will likely not substantially affect the pooled estimate. Therefore, the Committee did not exclude this RCT. For two RCTs with an open-label design it could not be judged to what extent this may have introduced bias (SHOT[49] and JELIS [23]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since CHD concerns a hard, clinical, non-subjective outcome. The authors of the JELIS trial furthermore noted that their trial was underpowered, which may be an explanation for the non-significant effects observed. However, as the number of cases in the Committee's pooled analysis (n=108) meet the criterion used to allow a conclusion with strong evidence (n=100), the Committee does not expect major concerns regarding the statistical power of this pooled analysis to demonstrate an effect (if there would actually be an effect).

4 Generalisability:
All RCTs (n=9) were performed in people with CHD. Therefore, the Committee could not evaluate whether or not effects of EPA and DHA supplementation on fatal CHD are similar in people with other types of ASCVD, such as stroke or PAD. In all RCTs, at least 80% of the study population comprised men. Whether effect modification by sex might be present could not be assessed.

3.2.7 Total MI

Based on the evaluation of RCTs, the Committee has concluded the following:
There is likely no effect of EPA and DHA supplementation of 1.4 to 4.8 grammes per day on the risk of total MI in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 7 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.4 to 4.8 g/d) on the risk of total MI in people with ASCVD.[21,23,38,43,46,48,49] In total, more than 100 events were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA/DHA supplementation on the risk of total MI (Table 2). There was moderate heterogeneity between studies (I^2=34%). Both neutral and protective effects were observed. Only one RCT (OPACH[46]) showed a reducing effect and the other RCTs did not show an effect of EPA/DHA supplementation on total MI. The heterogeneity might be (partially) due to the type of patients included in the RCTs: the OPACH trial included participants with (any type of) CVD (and were treated with chronic haemodialysis), whereas the other RCTs included participants with CHD. Based on visual inspection of the forest plot, the Committee assumes that the EPA and DHA dose, the type of intervention (EPA alone versus EPA plus DHA) and the proportion of statin users do likely not explain the heterogeneity. There were too few studies available to perform subgroup analyses in order to test these assumptions.

3 Considerations regarding the quality of the evidence:
For three RCTs with an open-label design it could not be judged to what extent this may have introduced bias (SHOT[49], JELIS [23] and Nosaka et al.[21]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since MI concerns a hard, clinical, non-subjective outcome. The authors of the JELIS trial furthermore noted that their trial was underpowered, which may be an explanation for the non-significant effects observed. However, as the number of cases in the Committee's pooled analysis (n=155) meet the criterion used to allow a conclusion with strong evidence

(n=100), the Committee does not expect major concerns regarding the statistical power of this pooled analysis to demonstrate an effect (if there would actually be an effect). According to the decision tree, in the case of no substantial heterogeneity and in the absence of major considerations, the evidence can be judged as strong.

4 Generalisability:
Almost all RCTs (n=6) were performed in people with CHD. Only the OPACH trial, the RCT that showed a statistically significant reducing effect, was performed in people with (any type of) CVD. Because there is no substantial heterogeneity between studies, the Committee sees no reason to expect that effects of EPA and DHA supplementation substantially differ between people with different types of ASCVD. In almost all RCTs, at least two-thirds of the study population comprised men. Whether results differed between men and women was not assessed in those studies and results were not presented for men and women separately. Because of this and because the number of MI events in women contributing to this analysis is likely rather small, the Committee could not conclude on whether there are indications to expect that associations are different in women compared to men.

3.2.8 Fatal MI

Based on the evaluation of RCTs, the Committee has concluded the following:
There is too little research to draw conclusions regarding the effect of EPA and DHA supplementation of >1 gramme per day on the risk of fatal MI in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 5 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.8 to 4.8 g/d) on the risk of fatal MI in people with ASCVD.[23,26,38,48,49] In total, 57 cases of fatal MI were reported. The number of cases reported is less than the number of cases required to draw a conclusion with strong evidence, including the conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of those 5 RCTs did not show an effect of EPA/DHA supplementation on the risk of fatal MI (Table 2) and no heterogeneity between studies was observed. However, those studies included too few cases to conclude that an effect is unlikely. Therefore, the Committee downgraded its conclusion and concluded that there is too little research to draw any conclusions.

3.2.9 Non-fatal MI

Based on the evaluation of RCTs, the Committee has concluded the following: **There is likely no effect of EPA and DHA supplementation of 1.6 to 4.8 grammes per day on the risk of non-fatal MI in people with ASCVD.**

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 8 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.6 to 4.8 g/d) on the risk of non-fatal MI in people with ASCVD.[21,23,26,38,43,45,48,49] In total, more than 100 events were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA/DHA supplementation on the risk of non-fatal MI (Table 2). There was moderate heterogeneity between studies (I^2=28%). Both neutral and protective effects were observed. Only one RCT (IEIS-4[26]) showed a statistically significant and substantial reducing effect of 48% on non-fatal MI. Th other RCTs did not show an effect of EPA/DHA supplementation on the risk of non-fatal MI. The subgroup analyses according to statin use do not suggest that the heterogeneity is (fully) explained by statin use since results do not substantially differ among the RCTs with a low proportion of statin users and those with a high proportion of statin users and because moderate heterogeneity remained in both subgroups. Furthermore, based on visual inspection of the forest plot, the Committee assumes that the EPA and DHA dose and the type of intervention (EPA alone versus EPA plus DHA) do likely not (fully) explain the heterogeneity either. To conclude, the Committee could not specify one factor that fully explains the heterogeneity observed and assumes that the moderate heterogeneity is most likely due to multiple differences between studies. The Committee can also not rule out that the heterogeneity is a result of the potential invalidity of the results of the RCT by Singh et al. (see point 3).

3 Considerations regarding the quality of the evidence:
There are some considerations regarding the validity of the results of Singh et al., who was suspected of research misconduct. The RCT by Singh et al. was the only study to show a substantial (statistically significant) 48% lower risk of non-fatal MI in the group receiving EPA/DHA supplements compared to the placebo group. Because of this, the Committee gave less weight to this MA in judging the totality of the evidence. For two RCTs with an open-label design it could not be judged to what extent this may have introduced performance bias (JELIS [23] and Nosaka et al.[21]). The lack of blinding might have resulted in performance bias, for example if

the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since MI concerns a hard, clinical, non-subjective outcome. The authors of the JELIS trial furthermore noted that their trial was underpowered, which may be an explanation for the non-significant effects observed in their study. However, as the number of cases in the Committee's pooled analysis (n=236) meet the criterion used to allow a conclusion with strong evidence (n=100), the Committee does not expect major concerns regarding the statistical power of this pooled analysis to demonstrate an effect (if there would actually be an effect). According to the decision tree, in the case of no substantial heterogeneity and in the absence of major considerations, the evidence can be judged as strong. The Committee notes the concerns with regard to the quality of the RCT by Singh et al, but disregarding this study leads to the same conclusion.

4 Generalisability:
All RCTs were performed in people with CHD. Because studies in people with other subtypes of ASCVD (stroke or PAD) were missing, the Committee could not evaluate whether the results observed also apply to these ASCVD subgroups.
In almost all RCTs, at least two-thirds of the study population comprised men. Whether results differed between men and women was not assessed in those studies and results were not presented for men and women separately. Because of this and because the total number of non-fatal MI events in women contributing to this analysis is likely rather small, the Committee could not conclude on whether there are indications to expect that associations are different in women compared to men.

3.2.10 Total stroke

Based on the evaluation of RCTs, the Committee has concluded the following: **There is inconclusive evidence regarding the effect of EPA and DHA supplementation of 1.1 to 4.8 grammes per day on the risk of total stroke in people with ASCVD.**

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:

There are 8 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.1 to 4.8 g/d) on the risk of total stroke in people with ASCVD.[21,38,40,42,43,45,46,48] In total, more than 100 events were reported. This is the first step required to mark

the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA/DHA supplementation on the risk of total stroke (Table 2). There was moderate to substantial heterogeneity between studies (I^2=47%), both in the direction and size/strength of the effects. RCTs showed predominantly neutral effects, but a single study showed a protective effect or a slight tendency towards an unfavourable effect, although not statistically significant. The pooled result was largely driven by the RCTs that evaluated EPA/DHA supplements with doses between 1 and 3 g/d (only 7 cases were reported among the RCTs with an EPA/DHA dose of ≥3 g/d compared to 132 cases among the RCTs with an EPA/DHA dose of 1-3 g/d). Based on visual inspection of the forest plot, the Committee saw no clear indication that effects differed according to EPA/DHA dose or statin use. Subgroups according to these factors became too small to perform subgroup analyses. The heterogeneity may be partially explained by the type of ASCVD patients. The analyses of the JELIS trial[40] contributing to this analysis were performed in solely stroke patients, whereas the other RCTs included people with CHD or total CVD. Excluding the JELIS trial reduced the I^2 to 36% (result NR). Since there was only one study in stroke patients, the hypothesis that the effect of EPA/DHA on stroke risk differs according to the type of ASCVD patient cannot be further explored. Overall, the Committee could not identify one factor that fully explains the heterogeneity observed and assumes that the heterogeneity is most likely due to multiple differences between studies. Since the heterogeneity can be considered substantial (I^2 is nearly 50%), the Committee found that no conclusion with strong evidence could be drawn. Therefore, it regarded the evidence as inconclusive.

3 Considerations regarding the quality of the evidence:
For two RCTs with an open-label design it could not be judged to what extent this may have introduced performance bias (JELIS[40] and Nosaka et al.[21]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The lack of blinding probably did not lead to detection bias, since stroke concerns a hard, clinical, non-subjective outcome. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
Five RCTs were performed in people with CHD,[21,38,43,45,48] two RCTs were

performed in people with total CVD[42,46] and one RCT was performed in people with stroke.[40] The RCTs in people with total CVD tend to show, by approximation, comparable results as compared to the RCTs in people with CHD (i.e. no effect). As explained under point 2, the Committee noted that the single study in stroke patients showed a reducing effect of EPA/DHA supplementation, which is not in line with the overall pooled result. However, as this observation is based on only one RCT, there is too little evidence to conclude on whether there are indications to expect associations are different in stroke patients compared to other ASCVD subgroups.

In almost all RCTs, at least two-thirds of the study population comprised men. Whether results differed between men and women was not assessed in those studies and results were not presented for men and women separately. Because of this and because the total number of stroke events in women contributing to this analysis is likely rather small, the Committee could not conclude on whether there are indications to expect that associations are different in women compared to men.

3.2.11 Sudden death

Based on the evaluation of RCTs, the Committee has concluded the following:

There is too little research to draw conclusions regarding the effect of EPA and DHA supplementation of >1 gramme per day on the risk of sudden death in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 5 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.8 to 3.5 g/d) on the risk of sudden death in people with ASCVD.[21,23,26,43,49] In total, 50 cases of sudden death were reported. The number of cases reported is less than the number of cases required to draw a conclusion with strong evidence, including the conclusion of 'an effect is unlikely' (at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of those five RCTs did not show an effect of EPA/DHA supplementation on the risk of sudden death (Table 2) and little heterogeneity between studies was observed (I^2=23%). However, those studies included too few cases to conclude that an effect is unlikely. Therefore, the Committee downgraded its conclusion and concluded that there is too little research to draw any conclusions.

3.2.12 Revascularisation

Based on the evaluation of RCTs, the Committee has concluded the following:

EPA and DHA supplementation of 1.4 to 4 grammes per day reduces the risk of revascularisation by an average of 27% in people with ASCVD. The evidence is strong.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 8 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.4 to 4 g/d) on the risk of revascularisation in people with ASCVD.[21,23,38,43,45-48] In total, nearly 1300 cases were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed). In these studies, revascularisation was defined as coronary revascularisation, including PCI and CABG.

2 Heterogeneity of the study findings:
A pooled analysis of those RCTs showed that EPA/DHA supplementation reduced the risk of revascularisation by an average of 27% (Table 2). No heterogeneity between studies was observed (I^2=0%). The Committee notes that the REDUCE-IT trial[47] contributed 42% to the pooled result. This RCT examined the effect of EPA alone, in a highly purified form (IPE), and found a significant reducing effect. The Committee sees, however, no convincing evidence that the reducing effect observed in the overall analysis is fully attributable to EPA, given the lack of heterogeneity between studies and because the results of all individual RCTs point towards reducing effects.

3 Considerations regarding the quality of the evidence:
For two RCTs with an open-label design it could not be judged to what extent this may have introduced performance bias (JELIS[40] and Nosaka et al.[21]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The Committee has noted no other major considerations regarding the quality of the RCTs that may impact the overall findings. According to the decision tree, in the case of no heterogeneity and in the absence of major considerations, the evidence is judged as strong.

4 Generalisability:
Six RCTs[21,23,38,43,45,48] included people with CHD and two RCTs[46,47] included people with (any type of) CVD. Based on visual inspection of the forest plot and given the fact that there is no heterogeneity between studies, the Committee

considered that there is no reason to expect that effects of EPA and DHA supplementation on revascularisation risk substantially differ between people with different types of ASCVD.
The Committee noted that in almost all RCTs at least two-thirds of the study population comprised men. Whether results differed between men and women was not assessed in those studies and results were not presented for men and women separately in those studies. Based on a comparison of studies with a smaller and greater proportion of women and considering that the absolute number of women in those studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.2.13 Angina pectoris

Based on the evaluation of RCTs, the Committee has concluded the following: **There is inconclusive evidence regarding the effect of EPA and DHA supplementation of 1.8 to 4.8 grammes per day on the risk of angina pectoris in people with ASCVD.**

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 6 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.4 to 4.8 g/d) on the risk of angina pectoris in people with ASCVD.[22,23,26,38,43,48] In total, 375 events were reported. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 100 cases are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs showed that EPA/DHA supplementation reduced the risk of angina pectoris by an average of 37% (Table 2). There was substantial heterogeneity between studies (I^2=56%). Both neutral effects and protective effects were observed. Also, substantial heterogeneity in the size of the effects was observed. The Committee noted that the majority of studies included in this analysis concern RCTs performed in a group of people of which only a small proportion used statins (<25%). The Committee saw no indication that statin use (fully) explains the heterogeneity as excluding the RCT performed in participants that almost all used statins,[22] did not reduce the level of heterogeneity (results NR). Based on visual inspection of the forest plot, the Committee saw also no clear indication that effects differed according to EPA/DHA dose or the type of the intervention (EPA and DHA combined versus EPA alone). Subgroups according to these factors became too small to perform subgroup analyses. To conclude, the Committee could not specify one factor that fully explains the heterogeneity

observed and assumes that the moderate heterogeneity is most likely due to multiple differences between studies. The Committee can also not rule out the possibility that the heterogeneity is a result of the potential invalidity of the results of the RCT by Singh et al.[26] (see point 3).

3 Considerations regarding the quality of the evidence:
There are some considerations regarding the validity of the results of Singh et al., who was suspected of research misconduct. The RCT by Singh et al. showed a substantial and statistically significant reducing effect of 70% on angina pectoris. Excluding this RCT, which contributed 20% to the pooled estimate, would probably result in a non-significant pooled effect. It is unknown how it would influence the level of heterogeneity. Given the above, the Committee gave less weight to this MA in judging the totality of the evidence. For one RCT with an open-label design it could not be judged to what extent this may have introduced performance bias (JELIS[40]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings. Because of the considerations with regard to the RCT by Singh et al. and since the heterogeneity between studies is substantial and no clear explanation could be found, the Committee found that no conclusion with strong evidence could be drawn. Therefore, it regarded the evidence as inconclusive.

4 Generalisability:
All RCTs were performed in people with CHD. Because studies in people with other subtypes of ASCVD (stroke or PAD) were missing, the Committee could not evaluate whether the results observed also apply to these ASCVD subgroups.
In almost all RCTs, at least 80% of the study population comprised men. Whether results differed between men and women was not assessed in those studies and results were not presented for men and women separately. Because of this and because the total number of female cases of angina pectoris contributing to this analysis is likely rather small, the Committee could not conclude on whether there are indications to expect that associations are different in women compared to men.

3.2.14 Heart failure

Based on the evaluation of RCTs, the Committee has concluded the following:
There is too little research to draw conclusions on the effect of EPA and DHA supplementation of >1 gramme per day on the risk of heart failure in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 3 RCTs that addressed the effect of EPA and DHA supplementation (range: 1.6 to 4.8 g/d) on the risk of heart failure in people with ASCVD.[21,38,45] This excludes a conclusion of 'an effect is unlikely' or a conclusion with strong evidence, for which at least 5 RCTs are required. Also, since in total 45 cases of heart failure were reported, a conclusion with limited evidence is not possible (for this, at least 3 RCTs and 60 cases are required).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA and DHA supplementation on the risk of heart failure (Table 2). There was no heterogeneity between studies. However, three RCTs is too few to draw a conclusion of 'likely no effect'. Therefore, the Committee downgraded its conclusions and concluded (based on the decision tree) that there is too little research.

3.2.15 Systolic blood pressure

Based on the evaluation of RCTs, the Committee has concluded the following:
There is likely no effect of EPA and DHA supplementation of 1.4 to 4.8 grammes per day on systolic blood pressure in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 6 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.4 to 4.8 g/d) on systolic blood pressure in people with ASCVD.[23,37-39,43,48] In total, nearly 1300 participants were included. This is the first step required to mark the evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 150 participants are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA/DHA supplementation on systolic blood pressure (Table 2). There was no heterogeneity between studies (I^2=0%).

3 Considerations regarding the quality of the evidence:
For two RCTs with an open-label design it could not be judged to what extent this

may have introduced performance bias (HEARTS[39] and JELIS[23]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in a slight underestimation or overestimation of the effect. The possibility that the outcome assessment has been affected cannot be ruled out. However, considering that no heterogeneity was observed between studies, the Committee assumes that this did not have substantially affected the overall pooled result. The Committee has noted no other major considerations regarding the quality of the RCTs that may impact the overall findings. According to the decision tree, in the case of no heterogeneity and in the absence of major considerations, the evidence can be judged as strong.

4 Generalisability:
All RCTs (n=6) were performed in people with CHD. Therefore, the Committee could not evaluate whether or not effects of EPA and DHA supplementation on systolic blood pressure are similar in people with other types of ASCVD, such as stroke or PAD.
In almost all RCTs, at least two-thirds of the study population comprised men (range: 68 to 100%). Whether results differed between men and women was not assessed in those studies and results were not presented for men and women separately. However, given the lack of heterogeneity between studies (studies differed with respect to the proportion of women included, amongst others) and considering that the absolute number of women in those studies is quite large (despite the relative share being low), the Committee sees no reason to expect that effects would be different in men and women.

3.2.16 LDL cholesterol

Based on the evaluation of RCTs, the Committee has concluded the following:
There is inconclusive evidence regarding the effect of EPA and DHA supplementation of 1.1 to 4.8 grammes per day on LDL cholesterol in people with ASCVD.

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 8 RCTs that addressed the effect of EPA/DHA supplementation (range: 1.1 to 4.8 g/d) on LDL cholesterol in people with ASCVD.[23,37-39,41,42,48,49] In total, nearly 2300 participants were included. This is the first step required to mark the

evidence as strong or to allow a conclusion of 'an effect is unlikely' (for which at least 5 RCTs and 150 participants are needed).

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA/DHA supplementation on LDL cholesterol (Table 2). There was substantial heterogeneity between studies (I^2=49%). Both neural effects and unfavourable effects were observed. The Committee also noted substantial heterogeneity in the size of the effect. Subgroup analyses according to proportion of statin users showed that EPA/DHA supplementation increased LDL cholesterol levels by an average of 0.29 mmol/L among the RCTs in which less than 25% of participants used statins, whereas no effect was observed among the RCTs in which nearly all participants used statins. The Committee noted, however, that in the subgroup of RCTs with a low proportion of statin users, substantial heterogeneity between studies remained. It therefore assumes that statin use does not fully explain the heterogeneity observed. The Committee furthermore noted that the strongest effects (i.e. the greatest changes in LDL cholesterol as well as statistically significant) were observed in the two RCTs with the shortest follow-up (1 and 4 months) and in which the participants were PAD patients.[37,41] This contrasts with the other RCTs, that generally showed smaller and non-statistically significant changes in LDL cholesterol, which included people with CHD or total CVD and had follow-ups of at least 1 year. The two RCTs showing an effect were moreover very small in number (33 and 29 participants), which prevented the Committee from performing subgroup analyses to further explore whether length of follow-up or patient type may influence the effect of EPA and DHA supplementation on LDL cholesterol. Furthermore, based on visual inspection of the forest plot, the Committee cannot rule out that higher doses of EPA and DHA may be more unbeneficial for the LDL cholesterol level (greater increase in LDL cholesterol) as compared to lower doses. Overall, the Committee identified multiple factors that might influence the effect of EPA and DHA supplementation on LDL cholesterol, but data are insufficient to identify which factor of factors are responsible for the substantial heterogeneity observed between studies. Therefore, the Committee found that no conclusion with strong evidence could be drawn and regarded the evidence as inconclusive.

3 Considerations regarding the quality of the evidence:
For two RCTs with an open-label design it could not be judged to what extent this may have introduced performance bias (JELIS[40] and HEARTS[39]). The lack of blinding might have resulted in performance bias, for example if the participants in the different groups behaved differently or received differential attention or care as a result of the intervention they were assigned to. This could, however, not be examined based on the data available. Therefore, the Committee cannot rule out the possibility that some performance bias was present and might have resulted in

a slight underestimation or overestimation of the effect. The Committee has noted no other concerns regarding the quality of the RCTs that may impact the overall findings.

4 Generalisability:
Five RCTs were performed in people with CHD,[23,38,39,48,49] one RCT was performed in people with total CVD[42] and two RCTs were performed in people with PAD.[37,41] The RCT in people with total CVD tend to show, by approximation, a comparable result as compared to the RCTs in people with CHD (i.e. no effect). As explained under point 2, the Committee noted that the two RCTs in PAD patients showed an increasing effect of EPA/DHA supplementation on LDL cholesterol, which is not in line with the overall pooled result (i.e. no effect). However, as this observation is based on only two very small RCTs, there is too little evidence to conclude on whether there are indications to expect associations are different in PAD patients compared to other ASCVD subgroups.
In almost all RCTs, at least two-thirds of the study population comprised men. Only the NAT2 trial[42] comprised of mostly women (35%). The result of this RCT was, by approximation, comparable to the overall effect (i.e. no effect). It was furthermore not assessed in the included studies whether results differed between men and women and results were not presented for men and women separately. Because of this and because the substantial heterogeneity observed between studies, the Committee could not conclude on whether there are indications to expect that associations are different in women compared to men.

3.2.17 Body weight

Based on the evaluation of RCTs, the Committee has concluded the following: **There is too little research to draw conclusions on the effect of EPA and DHA supplementation of >1 gramme per day on body weight in people with ASCVD.**

The following considerations were made by the Committee to come to this conclusion, following the steps of the decision tree:

1 Number of studies and cases:
There are 3 RCTs that addressed the effect of EPA and DHA supplementation (range: 2.3 to 4.8 g/d) on body weight in people with ASCVD.[38,39,48] This excludes a conclusion of ‘an effect is unlikely’ or a conclusion with strong evidence, for which at least 5 RCTs are required. However, since 3 RCTs with in total more than 90 participants were available, a conclusion with limited evidence is still possible.

2 Heterogeneity of the study findings:
A pooled analysis of these RCTs did not show an effect of EPA and DHA supplementation on body weight (Table 2). There was no heterogeneity between studies. Three RCTs is too few to draw a conclusion of ‘likely no effect’. Therefore, and because there was no obvious heterogeneity in direction of the effect, the

Committee downgraded its conclusions and concluded (based on the decision tree) that there is too little research.

3.2.18 Non-fatal CHD, non-fatal stroke and arrhythmia

The conclusions and accompanying explanation for the long-term health outcomes of non-fatal CHD, non-fatal stroke and arrythmia were largely similar. For the sake of readability, the description of the evaluation of these four outcomes is therefore combined.

Based on the evaluation of RCTs, the Committee concluded the following:
There is too little research to draw conclusions regarding the effect of EPA and DHA supplementation of >1 gramme per day on the risks of non-fatal CHD, non-fatal stroke and arrhythmia in people with ASCVD.

The following considerations were made by the Committee to come to these conclusions, following the steps of the decision tree:

1 Number of studies and cases:
There are two RCTs that addressed the effect of EPA and DHA supplementation of >1 g/d on the risk of non-fatal CHD in people with ASCVD (OFAMI[43] and JELIS[23]). The OFAMI trial[43] (cases n=70) showed no effect of EPA and DHA supplementation on non-fatal CHD (RR 1.30, 95%CI 0.81, 2.08), whereas the JELIS trial[23] (cases n=323) tended to show a reducing effect of 1.8 g/d of EPA on non-fatal CHD in people with CHD (HR 0.82, 95%CI 0.66, 1.02). One RCT addressed the effect on the risk of non-fatal stroke (Nosaka et al.[21]), and showed no effect of EPA supplementation of 1.8 g/d in people with CHD. Two RCTs addressed the risk of arrhythmia (OMEMI[45] and OFAMI[43]) in people with CHD. One RCT[43] showed no effect of EPA and DHA supplementation of 3.5 g/d, whereas the other RCT[45] tended to show an increasing effect of EPA and DHA supplementation of 1.6 g/d on arrhythmia (HR 1.84, 95%CI 0.98, 3.45). An evaluation of less than three studies provides too little evidence to draw conclusions on. Therefore, the Committee concluded that there was too little research regarding the effects of EPA and DHA supplementation on the health outcomes of non-fatal CHD, non-fatal stroke and arrhythmia in people with ASCVD.

3.3 Description of the selected RCTs

The individual RCTs from the selected MAs and the supplementary RCTs that the Committee selected for its evaluation are described below. For the RCTs that are included in an existing MA, the Committee used the data that are available in the MAs (and did, in principle, not consult the individual papers of these RCTs for additional

information). The characteristics and results of the RCTs are described in more detail in Annex C.

The multicentre, double-blind, placebo-controlled RCT named Alpha Omega was conducted by Kromhout et al. (2010).[28] The RCT had a 2-by-2 factorial design to examine the effects of EPA/DHA and alpha-linoleic acid (ALA) in people with a history of MI (up to 10 years before randomisation). EPA/DHA and ALA were administered through an enriched margarine. The Committee used the results of the two-way analyses in its evaluation (conform most MAs), meaning that the group receiving EPA/DHA and the group receiving EPA/DHA plus ALA were considered the intervention group and that the group receiving ALA and the control arm were considered the control group. For its evaluation of total CVD, the Committee used the results for major cardiovascular events, which Kromhout et al. defined as fatal and non-fatal cardiovascular events and the cardiac interventions PCI and CABG. The RCT did not show an effect of EPA/DHA-enriched margarine on total CVD, fatal CVD, total CHD, fatal CHD and total stroke. The authors has noted that the lack of an effect of EPA/DHA in this trial could be due to the fact that 85% of the participants were receiving statins and that the relatively low dose of EPA/DHA might be insufficient to demonstrate an effect on top of the lipid-lowering medication. The Committee has furthermore noted that relatively few cases of stroke were reported (n=21 in total), which might have contributed to a non-significant effect on total stroke.
Compliance with the intervention was assessed through measurements of fatty acid levels in plasma cholesteryl esters and telephone interviews, and 91% of the participants were found to adhere fully to the assigned intervention and to consume 20.6 g (± 2.8) margarine per day. According to the MAs by Abdelhamid et al. (2020)[24] and Rizos et al. (2012)[52], the risk of bias of the RCT was low. No notable concerns were raised. Funding was provided by the Netherlands Heart Foundation, the National Institutes of Health and Unilever R&D.

Gans et al. (1990)[37] observed no statistically significantly difference in change in LDL cholesterol between people who received fish oil supplements and people who received corn oil (control group) after 4 months. The study was performed in Dutch men and women with stable intermittent claudication (PAD).
The risk of bias of this RCT was assessed in the SR by Campbell et al. (2013)[53] and judged to be low. No concerns regarding the study quality were raised. It is unknown how LDL cholesterol was determined (with the Friedewald equation[54] or not). Also, it was not known if funding was received.

The large, multicentre, open-label GISSI-Prevenzione (GISSI-P) trial in people with recent MI (≤3 months) was conducted by Marchioli et al. (1999).[30] In origin, the trial was designed with a 2-by-2 factorial design, examining the effects of EPA/DHA and

vitamin E. In most MAs, the results of the two-way analyses were used, which means that the group receiving EPA/DHA and the group receiving EPA/DHA plus vitamin E were considered the intervention group and the group receiving vitamin E and the control arm the control group. Results of the intention-to-treat analyses showed that 0.88 g/d EPA and DHA supplementation (in a 1:2 ratio) reduced the risks of all-cause mortality, fatal CVD, total CHD, fatal CHD, fatal MI and sudden death (by 13 to 26%). No effects were observed on the risks of total CVD, non-fatal CVD, total MI, non-fatal MI, stroke, revascularisation, angina pectoris, arrhythmia or cancer.
Compliance with the intervention was assessed through capsule counts. After 12 months, 88% took the EPA/DHA supplements, and this was 71% after 42 months (end of the trial). The different MAs addressing this RCT judged the risk of bias as either moderate or high. This was mainly caused by a different judgement of the aspects of 'blinding of participants and personnel' and 'blinding of outcome assessors' and its potential consequences for the trial conclusions. The GISSI-P trial had an open-label design: participants, personnel and outcome assessors were not blinded. The Committee judged that this likely had little effect on the results, for the following main reasons. First, the health outcomes addressed in this RCT are hard, clinical outcomes, that are barely susceptible to (subjective) judgment. Second, although the open-label design might have led to participants in the control group changing their dietary habits, follow-up data from the GISSI-P trial indicate that the proportion of participants consuming ≥1 serving of fish per week was well-balanced across study groups at baseline and at 6 and 42 months of follow-up. Some uncertainty remains, however, because the exact amount of fish consumed (in g/d) was not reported and dietary intake was self-reported. The Committee also cannot rule out the possibility that any other behavioural changes (e.g. becoming more or less physically active, quitting smoking) occurred in either group as a result of the intervention the group was assigned to, which might have led to bias. Overall, the Committee judged the risk of bias in the GISSI-P trial as moderate. Funding was provided by multiple pharmaceutical companies (e.g. Pharmacia & Upjohn and Pfizer).
The Committee noted that this RCT started in an era in which statins were not commonly applied for the treatment of people with ASCVD. In the 21st century, the use of medication such as statins and antihypertensive drugs has increased significantly in people with CVD. As a result, the risk of a recurrent CVD reduces in these patients, which can make it more difficult to demonstrate an effect of EPA and DHA on top of the effects of these medications. Follow-up data of the GISSI-P trial clearly shows this trend in statin use: only 5% of the participants were on statin treatment at baseline (1993-1995), whereas this concerns 45% of the participants at the end of follow-up.

The HARP trial, conducted by Sacks et al. (1995),[38] did not show statistically significant effects of 4.8 g/d EPA and DHA supplementation on total CVD and fatal CVD in people with documented CHD. The Committee noted that the sample size was small (n=59 in

total) and that very few events occurred during the 28-month follow-up: 14 cases of total CVD were reported, of which only 1 case was fatal.
The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020).[24] They noted that there may have been selective reporting, as the study was registered retrospectively after publication. Compliance was not reported. Funding was provided by a pharmaceutical company (Warner Lambert-Parke Davis, now Pfizer). No other concerns were expressed by Abdelhamid or noted by the Committee.

In their open-label, parallel RCT named HEARTS, Alfaddagh et al. (2017)[39] did not observe an effect of 3.4 g/d EPA and DHA supplementation on the risk of all-cause mortality in people with stable CHD and on statin treatment. The Committee considered that this finding should be interpreted with caution, since very few participants (n=4) died during the 30-month follow-up. The number of events was much higher for total CVD and total CHD (n>75). However, no effect was demonstrated on those outcomes either.
The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020) and judged to be moderate to high.[24] Concerns were expressed regarding the open-label design, which might have introduced performance bias. The Committee considered that this is unlikely to have affected the outcome ascertainment, as the outcomes addressed in this RCT are hard, clinical, non-subjective outcomes. It cannot, however, be ruled out that systematic differences occurred between the intervention group and the control group (other than the intended EPA/DHA difference) as a consequence of the arm the participants were assigned to, for example differences in the participants dietary behaviour, dropout rate or compliance. Information on dietary intake and compliance is not available. With regard to the dropout rate, it was observed that the number of missings in the control group (n=28) was higher than in the intervention group (n=17), but reasons were not provided (or not known). Therefore, the Committee could not judge whether this might have introduced bias. Furthermore, Abdelhamid et al. judged that there may have been selective reporting, but that it might also be the case that not all results were published yet at the time that Abdelhamid et al. reviewed the literature (February 2019).[24]

The IEIS-4 trial, performed by Singh et al. (1997),[26] showed a statistically significant effect of 1.8 g/d EPA/DHA supplementation on all-cause mortality and non-fatal CVD: the risk of all-cause mortality reduced by 50% and the risk of non-fatal CVD by 56%. An important difference between this RCT and many other RCTs is that Singh et al. used aluminium hydroxide as comparator whereas most others used either an oil (e.g. corn oil, olive oil, sunflower oil) or no placebo. It is furthermore of interest to note that all participants were men.
The Committee noted that the main author, Singh, is under suspicion of research misconduct,[27] and it has therefore interpreted the results of this RCT with caution. The

risk of bias of this RCT was assessed in the MAs by Rizos et al. (2012).[52] They did not note any other concerns regarding the quality of this study. The funding source is unknown.

Yokoyama et al. (2007)[23] performed an open-label, blinded endpoint-evaluation RCT named JELIS to examine the effect of 1.8 g EPA supplementation, in addition to statin treatment, on coronary events in Japanese participants with hypercholesteremia (LDL cholesterol level >4.4. mmol/L). No other antihyperlipidaemic drugs were allowed for the duration of the trial. For the current evaluation, the Committee used the results from the subgroup analysis in people with documented CHD. The RCT showed that EPA supplementation reduced the risk of total CVD by 21%. A borderline significant reducing effect of 19% was observed on total CHD. No statistically significant effects were observed on fatal CHD and non-fatal CHD, although the effect estimates were similar to total CHD. Yokoyama et al. noted that the trial was underpowered for subgroup analyses (as described here), which may be an explanation for the non-significant effects observed for fatal CHD and non-fatal CHD. The authors also noted that the average intake of fish is generally high in Japanese, in whom the effect of EPA may be more pronounced than in those with a low fish intake.
Compliance with the statins until termination of the trial (after a mean of 4.6 years) was comparable in the control group and the intervention group (~74%). Compliance with the EPA supplements was 71%. The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020).[24] Concerns were expressed regarding the open-label design, which might have led to performance bias. However, based on the data available, it could not be judged if this was the case in this RCT. Funding was provided by a pharmaceutical company (Mochida Pharmaceutical Co.). No other concerns regarding the quality of the study were noted.

Mori et al. (1992)[41] performed a RCT in Australian men with symptomatic and angiographically demonstrated peripheral artery disease. They observed a significant increase in LDL cholesterol in the group receiving fish oil supplements (total dose of EPA and DHA of 4.6 g/d) as compared to the control group receiving olive oil after 10 weeks.
The risk of bias of this RCT was assessed in the SR by Campbell et al. (2013)[53] and judged to be low. No concerns regarding the study quality were raised. The Committee has noted that LDL cholesterol was determined using the Friedewald equation.[54] LDL cholesterol determined with the Friedewald equation is considered less accurate in people with hypertriglyceridaemia (triglyceride levels ≥400 mg/d or 4.5 mmol/L). In the current study, average triglyceride levels at baseline were 2.3 ± 0.4 mmol/L and 2.8 ± 0.4 mmol/L in the intervention group and the control group (well below 4.5 mmol/L), respectively, and not statistically significantly different. Also, triglyceride levels changed little during the follow-up. Because of this, the Committee assumed that using the

Friedewald equation had not introduced substantial bias. Compliance was determined by capsule count and found to be good: 99% and 98% of the capsules were taken in the intervention group and the control group, respectively. Funding was provided by the National Heart Foundation of Australia.

In the NAT2 trial,[42] Souied et al. (2013) did not observe effects of supplementation with 1.1 g/d of EPA and DHA, as compared to olive oil, on the risks of all-cause mortality and total stroke in French people with CVD and early age-related macular degeneration.
Compliance with the supplements was assessed from unused capsules and serum PUFA levels. The overall compliance over the 3 years was moderate, but comparable in the intervention group and the control group: 69% and 71%, respectively. The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020) and judged to be low.[24] No concerns regarding the study quality were raised. Funding was provided by a pharmaceutical company (Laboratoire Chauvin).

Nosaka et al. (2017)[21] examined the effect of EPA (only) in statin-treated people who had a PCI after acute MI. EPA supplementation started within 24 hours after PCI. EPA supplementation had a borderline significant reducing effect of 78% on the risk of all-cause mortality. No effects on fatal CVD, non-fatal MI and sudden death were observed. The Committee considered that these results should be interpreted with caution, especially the results for non-fatal MI and sudden death, as very few events occurred, e.g. only one non-fatal MI event was reported in the intervention group and none in the control group.
The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020).[24] Concerns were expressed with regard to the open-label design. Also, it could not be judged whether results had been selectively reported, as the study was registered after data collection was completed. No information on compliance was reported. Funding was provided by a pharmaceutical company (Mochida Pharmaceutical Co.).

The Nutristroke trial was performed by Garbagnati et al. (2009).[31] Garbagnati et al. did not observe an effect of 0.5 g/d EPA and DHA supplementation on the risks of total CVD or fatal CVD in people with stroke. It is important to note that the number of events was very low: zero (fatal) CVD cases were reported in the intervention group and four in the control group.
The risk of bias was assessed by Abdelhamid et al. (2020).[24] They reported that the dropout rate was high, compliance was unclear and selective reporting could not be judged. Funding was provided by Sigma-Tau Health Science (part of a pharmaceutical company).

Nye et al. (1990)[22] examined the effect of EPA supplementation (Maxepa) in a dose of 2.2 g/d as compared to olive oil capsules in people undergoing PTCA. They found no effect on total CHD risk.
Compliance with the intervention was not reported. The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020) and judged to be low.[24] No concerns regarding the study quality were raised. Funding was provided by a pharmaceutical company (Scherer Ltd).

In its double-blind RCT named OFAMI, Nilsen et al. (2001)[43] did not observe an effect of 4 g/d EPA/DHA supplementation on the risks of fatal CVD, non-fatal CVD, total CHD, non-fatal CHD and non-fatal MI in people with an acute MI.
Participants were from Norway, where fish intake is relatively high compared to many other (European) countries. The authors noted that the participants' baseline EPA and DHA intake may have been higher than in many other studies. It is possible that the optimal cardioprotective dose of EPA and DHA had already been reached and that any additional amount of EPA or DHA did not further affect the cardiovascular risk. This hypothesis is in contrast to Yokoyama et al., who suggested that the effect of EPA and DHA would be more pronounced when baseline EPA/DHA intake is higher.
The Committee has noted that the number of participants using statins increased during the trial, but that the increase was comparable in the intervention group and the control group: 6% of the intervention group and 8% of the control group used statins before admission for acute MI, 42% and 45% at hospital discharge and 71% and 65% after 12 months, respectively. Although the proportion of statin users was comparable after 12 months, it is unknown if the time of introduction of those statins was also comparable in both arms. Therefore, the Committee cannot judge to what extent this might have affected the results.
Compliance rates of 82% and 86% were reported in the intervention group and the control group, respectively. The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020).[24] Due to missing data on dropouts, it was unclear whether attrition bias might have played a role. Also, it could not be judged whether results had been selectively reported, as the study was registered retrospectively. The study was supported by pharmaceutical companies (Pharmacia & Upjohn and Pronova).

In the OMEGA trial, Rauch et al. (2010)[32] performed per-protocol analyses in 3804 of the 3851 recruited people with an acute MI (dropout: 1%). The RCT did not show an effect of 1-year EPA/DHA supplementation on the risks of fatal CVD, total CHD and fatal CHD.
Compliance with the intervention was assessed through pill counts. In both the intervention group and the control group, 93% of the participants took at least 70% of the prescribed capsules. The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020) and judged to be low.[24] No concerns regarding the study

quality were raised. Funding was provided by a pharmaceutical company (Tromsdorff Arzneimittel).

The multicentre, double-blind, placebo-controlled parallel OMEGA-REMODEL trial was performed by Heydari et al. (2016).[44] It was conducted in people with acute MI to examine the effect of 6-month EPA/DHA supplementation (compared to corn oil) on the risk of all-cause mortality. All participants received lifestyle counselling, including dietary advise for standard post-MI care, but no specific recommendations were given regarding dietary intake of EPA and DHA. After a median follow-up of 24 months, 8 participants in the intervention group and 3 participants in the control group had died. This difference was not statistically significant (P=0.22). The Committee noted that relatively few cases were reported, which may explain the lack of a statistically significant effect.
Compliance was assessed through pill counts and found to be 96% in both the intervention group and the control group. The risk of bias of this RCT was assessed in the MA by Popoff et al.[55] according to a modified version of the Cochrane Risk of Bias Tool. Popoff et al. judged the risk of bias as low.

Recently, the multicentre, double-blind OMEMI trial was conducted by Kalstad et al. (2021).[45] It was performed in older adults with an acute MI (2-8 weeks). The Committee used the results for MACE for the evaluation of total CVD. Kalstad et al. defined MACE as a composite of non-fatal MI, unscheduled revascularisation, stroke, hospitalisation for heart failure or all-cause mortality. Results of the intention-to-treat analyses did not show an effect of EPA and DHA supplementation on the risk of all-cause mortality or total CVD. Per-protocol analyses yielded similar results. The effect on total CVD did not differ based on age, sex, body mass index (BMI), diabetes, previous MI, previous heart failure, previous hyperlipidaemia, levels of triglycerides or use of n-3 PUFA supplement at baseline. Effect modification was not examined for all-cause mortality.
Adherence to the intervention was assessed via an interview and measurements of serum EPA and DHA concentrations. Self-reported adherence, defined as no more than four consecutive weeks without taking the prescribed supplement, was 88%. Median serum EPA concentrations increased by 87% and serum DHA concentration by 16% in the intervention group after 2 years (relative changes from baseline). In the control group, serum EPA and DHA concentrations changed by -13% and -8%, respectively. The Committee judged the compliance with the intervention as good. Based on a risk of bias assessment that the Committee performed using the Cochrane collaboration tool RoB2, the overall risk of bias of this RCT was judged as low.
The Committee noted that the definition of MACE also included all-cause mortality, whereas all-cause mortality may also include non-CVD-related events. Furthermore, it must be noted that the authors added hospitalisation for heart failure to the definition of MACE (by protocol amendment) while recruitment was still ongoing. This decision was

made by the steering group and before unblinding, so the Committee expects this had not affected the quality of the study.

In the OPACH trial, conducted by Svensson et al. (2006),[46] a statistically significant effect of 1.7 g/d of fish oil, as compared to olive oil, was observed on the risk of total CHD in people who had established CVD and were treated with chronic haemodialysis. No effect was observed on revascularisation.
The risk of bias of this RCT was assessed in the MA by Rizos et al. (2012),[52] who did not note any concerns regarding the quality of this study. Funding was provided by both industry and non-industry parties.

The multicentre, multi-country, double-blind ORIGIN trial was performed in men and women aged 50 years and over who had prediabetes or diabetes and a high CVD risk. For the current evaluation, the Committee used the results from the publication by Dagenais et al. (2019),[33] who performed analyses in the subgroup of people with PAD. In this analysis, the effect of 0.84 g/d of EPA and DHA supplementation, as compared to olive oil supplementation, on the risk of PAD progression was examined. Dagenais et al. defined PAD progression as the occurrence of either a decrease of at least 0.1 unit in ankle-brachial index (ABI) as compared to baseline ABI (asymptomatic PAD) or claudication with an ABI of 0.90 or less or lower limb revascularisation or amputation as the result of arterial disease (symptomatic PAD). No effects of EPA and DHA supplementation were observed on PAD progression. There was no difference in risk of asymptomatic PAD or symptomatic PAD.
The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020) and judged to be low.[24] A limitation might be that the trial was sponsored by Sanofi Aventis (pharmaceutical company), and any potential role of the funder in the design, analysis or reporting of the study is unknown.

Bhatt et al. (2019)[20] performed the multicentre, multi-country, double-blind REDUCE-IT trial to examine the effect of 4 g/d of IPE (a highly purified EPA ethyl ester) compared to a placebo (mineral oil) on the risk of ischemic events in people with documented CVD or with diabetes and other risk factors, who received statin therapy, had a fasting triglyceride level of 135 to 499 mg/dL (the majority had elevated triglyceride levels) and had an LDL cholesterol level of 41 to 100 mg/dL. The subgroup analysis in people with documented CVD was used in the Committee's evaluation. A publication by Peterson et al. (2021)[47] reported on the revascularisation outcome. It was shown that EPA supplementation significantly reduced the risk of total CVD by 27% and the risk of coronary revascularisation by 37%. Total CVD was defined as cardiovascular death, non-fatal MI, non-fatal stroke, coronary revascularisation or unstable angina. Excluding coronary revascularisation and unstable angina from the definition of total CVD gave the same result.

Measurements of serum EPA were used to assess compliance. Serum EPA rose from 26 ug/mL at baseline to 144 ug/mL in the intervention group and slightly fell to 23 ug/mL in the control group after 1 year, suggesting good compliance during the first period of the study. Levels of serum EPA were, however, not reported after 6.2 years (end of trial). The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020).[24] No notable concerns regarding allocation, blinding, attrition or reporting were raised.
According to the Committee, strengths of this RCT include the relatively long supplementation duration and follow-up (median: 4.9 years), the large sample size and the low dropout rate (<1% loss over 6.2 years of follow-up). A limitation might be that the trial was sponsored by Amarin Pharma (supplement manufacturer), and any potential role of the funder in the design, analysis or reporting of the study is unknown.

The Committee has noted that this potential limitation not only pertains to this RCT: almost all RCTs received funding from industry (often pharmaceutical) companies. Another limitation concerns the placebo used, as further described below.
Bhatt et al. themselves noted as a limitation that the mineral oil that was used as placebo might have limited the absorption of the background statin therapy. This might be an explanation for the observed increase in LDL cholesterol in the control group, while only a very small change in LDL cholesterol was seen in the intervention group. As a consequence, this might have strengthened the observed CVD-reducing effect of IPE supplementation. The authors assumed, however, that the (relatively small) difference in LDL cholesterol between the intervention group and the control group would likely not explain the 27% lower CVD risk observed. The use of the mineral oil as placebo was also mentioned as a (main) potential limitation of REDUCE-IT by other researchers.[56-59] Mineral oils may not only reduce the absorption of statins, but also of other drugs, and may raise levels of inflammatory markers such as C-reactive protein. In fact, in REDUCE-IT a considerable rise in high-sensitivity C-reactive protein was observed in the control group. In this context, it is of particular interest that those (substantial) increases in LDL cholesterol and C-reactive protein where not observed in the STRENGTH trial.[50] In STRENGTH, also performed in CVD patients who had elevated triglyceride levels and received statin therapy, corn oil was used as placebo. In the corn oil group, both LDL cholesterol and C-reactive protein levels slightly decreased, contrary to the effects exerted by the mineral oil in REDUCE-IT. Moreover, the STRENGTH trial did not confirm the large CVD-reducing effects of IPE observed in REDUCE-IT. In fact, STRENGTH did not show an effect of EPA and DHA supplementation on total CVD in ASCVD patients. Like STRENGTH, also several other (previous) RCTs did not confirm the results of REDUCE-IT. What exactly explains the contrasting results is not yet fully understood. Researchers suggest that, besides the type of placebo (mineral oil), also the specific supplement used in REDUCE-IT, namely IPE (EPA in a highly purified form), might explain the different findings between

REDUCE-IT and most other RCTs, which mainly investigated a supplement containing (less purified) EPA and DHA. Other potential explaining factors might include dose or baseline ASCVD risk.[56,57,59]

The SHOT trial was conducted by Eritsland et al. (1996).[49] This parallel RCT was designed with a 2-by-2 factorial design to examine the effects of fish oil and warfarin. For statistical analysis, Eritsland et al. considered the group receiving fish oil and the group receiving fish oil plus warfarin as the intervention group and the group receiving warfarin and the control arm (no treatment) as the control group. No statistically significant effect of 3.3 g/d of fish oil was observed on the risks of all-cause mortality, total CVD, fatal CVD, total CHD and fatal CHD in people admitted for CABG. Neither was an effect found on change in LDL cholesterol. The Committee noted that few events occurred during the 1-year follow-up: from 11 cases of fatal CHD to 27 cases of total CVD. This might explain the lack of statistical significance. The Committee has furthermore noted that the majority of participants were men (87%).
Capsule count and measurements of serum EPA and DHA were used to assess compliance. On average, 88% of capsules were taken. Serum EPA and DHA rose in the intervention group and slightly fell in the control group after nine months. Together, this suggests generally good compliance with the intervention.
The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020).[24] Concerns were expressed regarding the open-label design, which might have led to performance bias. However, Eritsland et al. noted that participants were not aware of their assignment. Therefore, and because the outcomes addressed concern hard, clinical outcomes, the Committee expects that the open-label design will not have substantially affected (biased) the results. Funding was provided by pharmaceutical companies (Pronova and Nycomed Pharma). No other concerns regarding the quality of the study were noted.

Recently, the large multicentre, multi-country, double-blind STRENGTH trial in statin-treated people with established ASCVD was conducted by Nicholls et al. (2020).[50] The Committee has used the results for MACE for its evaluation of total CVD. Nicholls et al. defined MACE as cardiovascular death, non-fatal MI, non-fatal stroke, coronary revascularisation and hospitalisation for unstable angina. No effects of 4 g/d of EPA and DHA supplementation, as compared to corn oil, were observed on the risks of all-cause mortality, total CVD and fatal CVD. However, the trial showed that EPA/DHA supplementation significantly reduced the risk of total CHD by 15%.
The authors of the trial reported that EPA and DHA were administered as carboxylic acid formulation, which has a greater bioavailability than ethyl esters (frequently used in other trials, such as the OMEGA trial and the JELIS trial) and may explain heterogeneity between studies. The Committee noted that many participants were overweight (mean BMI: 32 ± 6 kg/m^2).

An important notion is that this trial was terminated prematurely (after a median of 42 months instead of an intended duration of 60 months) due to futility and the probability of an increased, albeit small, risk of atrial fibrillation in the EPA/DHA group. The authors assumed that the early termination of the study did not raise concerns regarding the power of the study (the number of events reported so far were close to the original sample size calculations). Based on a risk of bias assessment that the Committee performed using the Cochrane collaboration tool RoB2, the Committee has no other concerns related to the quality of this study and has judged the overall risk of bias of this RCT as low.

Galan et al. (2010)[34] conducted the double-blind, parallel RCT named SU.FOL.OM3, which was designed with a 2-by-2 factorial design to examine the effects of EPA/DHA and B-vitamins on health outcomes in patients with a history of ischaemic heart disease or stroke. For statistical analysis, Galan et al. considered the group receiving EPA/DHA and the group receiving EPA/DHA plus B-vitamins as the intervention group and the group receiving B-vitamins and the control arm as the control group. Results of the intention-to-treat analyses did not show an effect of EPA and DHA supplementation on the risk of any of the health outcomes evaluated. For fatal CHD, few events were reported during the 4-year follow-up: n=5 in the intervention group and n=13 in the control group. This might explain why statistical significance was not reached (HR 0.38, 95%CI 0.14-1.07).
The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020) and judged to be low.[24] No concerns regarding the study quality were raised. According to the Committee, strengths of this RCT include the relatively long supplementation duration and follow-up (median: 4.7 years), the low dropout rate (10% loss over 4 years of follow-op) and the high compliance (86% in all groups). A limitation might be that funding was provided by industry companies (e.g. Unilever, Roche and Merck) and any potential role of the funders in the design, analysis or reporting of the study is unknown.

In their SCIMO trial, Von Schacky et al. (1999)[48] observed no effect of 2.3 g/d EPA and DHA supplementation on CVD-, CHD- and stroke outcomes, all-cause mortality and revascularisation in people with CHD. Although the point estimates suggested a strong reducing effect (of about 70%), the results were far from statistically significant. This might be due to the relatively small sample size of the study (n=233 participants) and very few events reported. For example, the number of events in the intervention group and the control group was: 2 and 1 for all-cause mortality, 2 and 6 for non-fatal CVD, 1 and 3 for non-fatal MI and 1 and 3 for revascularisation, respectively.
Based on measurements of erythrocyte phospholipids during follow-up, compliance was considered good: levels increased on average from 4.6% at baseline to 11.8% at 24 months in the intervention group and did not alter in the control group. The risk of bias of this RCT was assessed in the MA by Abdelhamid et al. (2020).[24] Due to missing

data on dropouts, it was unclear whether attrition bias might have played a role. Also, it could not be judged whether results had been selectively reported, as no register or study protocol was available. Capsules and funding were provided by Pronova (medical company), but it was stated that they had no role in analysis or publication. No other concerns regarding the quality of the study were noted.

3.4 Summary of conclusions

The Committee's conclusions regarding effects of EPA and DHA supplementation of ≤1 gramme per day and of >1 gramme per day with health outcomes in people with ASCVD are summarised in Table 3.

Table 3 Overview of conclusions regarding effects of EPA and DHA supplementation on health outcomes in people with ASCVD, based on RCTs

Health outcome	Conclusion for EPA/DHA supplementation of ≤1 g/d	Conclusion for EPA/DHA supplementation of >1 g/d
All-cause mortality	Likely no effect	Likely no effect
Total CVD	Likely no effect	EPA plus DHA: likely no effect EPA alone: limited evidence for a reducing effect
Fatal CVD	Likely no effect	Likely no effect
Non-fatal CVD	Too little research	Too little research
Total CHD	Too little research	Strong evidence for a reducing effect
Fatal CHD	Limited evidence for a reducing effect	Likely no effect
Non-fatal CHD	N/A	Too little research
Total MI	Too little research	Likely no effect
Fatal MI	Too little research	Too little research
Non-fatal MI	Too little research	Likely no effect
Total stroke	Too little research	Inconclusive evidence
Non-fatal stroke	N/A	Too little research
Sudden death	Limited evidence for a reducing effect	Too little research
Revascularisation	Too little research	Strong evidence for a 27% reducing effect
Angina pectoris	Too little research	Inconclusive evidence
Heart failure	Too little research	Too little research
Arrhythmia	Too little research	Too little research
PAD progression	Too little research	N/A
Depression	Too little research	N/A
Cancer	Too little research	N/A
Systolic blood pressure	Too little research	Likely no effect
LDL cholesterol	Too little research	Inconclusive evidence
Body weight	N/A	Too little research

Abbreviations: ASCVD: atherosclerotic cardiovascular disease; CHD: coronary heart disease; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; LDL: low-density lipoprotein; MI: myocardial infarction; PAD: peripheral arterial disease; RCT: randomised controlled trial.

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Annexes

Annex A Search strategy and study selection

A.1 Search strategy for systematic reviews including meta-analyses

The Committee performed a literature search to identify relevant systematic reviews (SRs) including meta-analyses (MAs) on the relationship between EPA and/or DHA intake and health outcomes in people with atherosclerotic cardiovascular disease (ASCVD). Publications were searched in PubMed and Scopus on 8th July 2021 using the following search strategies:

PubMed

(Coronary disease [MeSH] OR Acute coronary syndrome [MeSH] OR Angina pectoris [MeSH] OR Coronary artery disease [MeSH] OR Myocardial infarction [MeSH] OR Peripheral arterial disease [MeSH] OR Intermittent claudication [MeSH] OR Stroke [MeSH] OR Brain ischemia [MeSH] OR Cerebrovascular disorders [MeSH] OR Percutaneous coronary intervention [MeSH] OR Coronary artery bypass [MeSH] OR Coronary disease [TIAB] OR Coronary heart disease [TIAB] OR Acute coronary syndrome [TIAB] OR Angina pectoris [TIAB] OR Angina [TIAB] OR Ischemic heart disease [TIAB] OR Ischaemic heart disease [TIAB] OR Coronary artery disease [TIAB] OR Coronary Arteriosclerosis [TIAB] OR Myocardial infarction [TIAB] OR Heart attack [TIAB] OR Peripheral arterial disease [TIAB] OR Peripheral vascular disease [TIAB] OR Intermittent claudication [TIAB] OR Stroke [TIAB] OR Acute stroke [TIAB] OR Cerebrovascular Apoplexy [TIAB] OR Apoplexy [TIAB] OR Ischemic stroke [TIAB] OR Ischaemic stroke [TIAB] OR Hemorrhagic stroke [TIAB] OR Haemorrhagic stroke [TIAB] OR Cerebrovascular accident [TIAB] OR Acute cerebrovascular accident [TIAB] OR Cerebrovascular stroke [TIAB] OR Brain vascular accident [TIAB] OR Brain ischemia [TIAB] OR Cerebral ischemia [TIAB] OR Cerebral stroke [TIAB] OR Brain accident [TIAB] OR Brain infarction [TIAB] OR Cerebral infarction [TIAB] OR Transient ischemic attack [TIAB] OR TIA [TIAB] OR Cerebrovascular* [TIAB] OR Subarachnoid haemorrhage [TIAB] OR Intracerebral hemorrhage [TIAB] OR Intracranial hemorrhages [TIAB] OR Coronary revascularization [TIAB] OR Percutaneous coronary intervention [TIAB] OR Coronary artery bypass graft surgery [TIAB] OR Percutaneous transluminal coronary angioplasty [TIAB] OR Percutaneous transluminal angioplasty [TIAB] OR Coronary angioplasty [TIAB] OR Atherosclerotic cardiovascular disease [TIAB] OR Carotid artery disease [TIAB] OR CHD [TIAB] OR ACS [TIAB] OR IHD [TIAB] OR CAD [TIAB] OR MI [TIAB] OR AMI [TIAB] OR PAD [TIAB] OR CVA [TIAB] OR CVAs [TIAB] OR TIA [TIAB] OR PCI [TIAB] OR CABG [TIAB] OR PTCA [TIAB] OR PTA [TIAB] OR ASCVD [TIAB])

AND

("Fatty Acids, Omega-3"[MeSH] OR omega-3 fatty acid* [TIAB] OR omega 3 fatty acid* [TIAB] OR n-3 fatty acid* [TIAB] OR n3 fatty acid* [TIAB] OR n 3 fatty acid* [TIAB] OR omega-3 [TIAB] OR omega 3 [TIAB] OR n-3 polyunsaturated [TIAB] OR n3 polyunsaturated [TIAB] OR n 3 polyunsaturated [TIAB] OR n-3 PUFA [TIAB] OR n3 PUFA [TIAB] OR Docosahexaenoic acid* [TIAB] OR docosahexenoic acid* [TIAB] OR eicosapentaenoic acid* [TIAB] OR eicosapentenoic acid* [TIAB] OR DHA [TIAB] OR EPA [TIAB] OR fish fatty acid* [TIAB] OR Fish oils [MeSH] OR Fish oil* [TIAB] OR marine oil* [TIAB] OR algal oil* [TIAB] OR Fishes [MesH] OR Fishes [TIAB] OR Fish [TIAB] OR seafood [MeSH] OR seafood [TIAB])

AND

(Systematic review[publication type] OR Meta-analysis[publication type] OR Review Literature as Topic [MesH] OR review[tiab] OR Meta-Analysis[MesH] OR "meta-analysis"[tiab] OR meta analysis [tiab] OR metaanalysis[tiab] OR quantitative review[tiab] OR quantitative overview[tiab] OR Systematic Reviews as Topic [MesH] OR systematic review[tiab] OR systematic overview[tiab] OR methodologic review[tiab] OR methodologic overview[tiab] OR individual participant data [TIAB] OR individual patient data [TIAB] OR IPD [TIAB] OR individual-level data [TIAB] OR pooled analysis [TIAB] OR pooled analyses [TIAB] OR multi-center study [TIAB] OR multi-cohort study [TIAB])

Limit: from 2000

Scopus

TITLE-ABS("Coronary disease") OR TITLE-ABS("Acute coronary syndrome") OR TITLE-ABS("Angina pectoris") OR TITLE-ABS("Coronary artery disease") OR TITLE-ABS("Myocardial infarction") OR TITLE-ABS("Peripheral arterial disease") OR TITLE-ABS("Intermittent claudication") OR TITLE-ABS(Stroke) OR TITLE-ABS("Brain ischemia") OR TITLE-ABS("Cerebrovascular disorders") OR TITLE-ABS("Percutaneous coronary intervention") OR TITLE-ABS("Coronary artery bypass") OR TITLE-ABS("Coronary heart disease") OR TITLE-ABS(Angina) OR TITLE-ABS("Ischemic heart disease") OR TITLE-ABS("Ischaemic heart disease") OR TITLE-ABS("Coronary Arteriosclerosis") OR TITLE-ABS("Heart attack") OR TITLE-ABS("Peripheral vascular disease") OR TITLE-ABS("Acute stroke") OR TITLE-ABS("Cerebrovascular Apoplexy") OR TITLE-ABS(Apoplexy) OR TITLE-ABS("Ischemic stroke") OR TITLE-ABS("Ischaemic stroke") OR TITLE-ABS("Hemorrhagic stroke") OR TITLE-ABS("Haemorrhagic stroke") OR TITLE-ABS("Cerebrovascular accident") OR TITLE-ABS("Acute cerebrovascular accident") OR TITLE-ABS("Cerebrovascular stroke") OR TITLE-ABS("Brain vascular accident") OR TITLE-ABS("Brain ischemia") OR TITLE-ABS("Cerebral ischemia") OR TITLE-

ABS("Cerebral stroke") OR TITLE-ABS("Brain accident") OR TITLE-ABS("Brain infarction") OR TITLE-ABS("Cerebral infarction") OR TITLE-ABS("Transient ischemic attack") OR TITLE-ABS(TIA) OR TITLE-ABS(Cerebrovascular*) OR TITLE-ABS("Subarachnoid haemorrhage") OR TITLE-ABS("Intracerebral hemorrhage") OR TITLE-ABS("Intracranial hemorrhages") OR TITLE-ABS("Coronary revascularization") OR TITLE-ABS("Percutaneous coronary intervention") OR TITLE-ABS("Coronary artery bypass graft surgery") OR TITLE-ABS("Percutaneous transluminal coronary angioplasty") OR TITLE-ABS("Percutaneous transluminal angioplasty") OR TITLE-ABS("Coronary angioplasty") OR TITLE-ABS("Atherosclerotic cardiovascular disease") OR TITLE-ABS("Carotid artery disease") OR TITLE-ABS(CHD) OR TITLE-ABS(ACS) OR TITLE-ABS(IHD) OR TITLE-ABS(CAD) OR TITLE-ABS(MI) OR TITLE-ABS(AMI) OR TITLE-ABS(PAD) OR TITLE-ABS(CVA) OR TITLE-ABS(CVAs) OR TITLE-ABS(TIA) OR TITLE-ABS(PCI) OR TITLE-ABS(CABG) OR TITLE-ABS(PTCA) OR TITLE-ABS(PTA) OR TITLE-ABS(ASCVD)

AND

TITLE-ABS("Fatty Acids, Omega-3") OR TITLE-ABS("omega 3 fatty acid*") OR TITLE-ABS("n-3 fatty acid*") OR TITLE-ABS("n3 fatty acid*") OR ("n 3 fatty acid*") OR TITLE-ABS(omega-3) OR TITLE-ABS("omega 3") OR TITLE-ABS("n-3 polyunsaturated") OR TITLE-ABS("n3 polyunsaturated") OR TITLE-ABS("n 3 polyunsaturated") OR TITLE-ABS("n-3 PUFA") OR TITLE-ABS("n3 PUFA") OR TITLE-ABS("Docosahexaenoic acid*") OR TITLE-ABS("Docosahexenoic acid*") OR TITLE-ABS("Eicosapentaenoic acid*") OR TITLE-ABS("Eicosapentenoic acid*") OR TITLE-ABS(DHA) OR TITLE-ABS(EPA) OR TITLE-ABS("fish fatty acid*") OR TITLE-ABS("Fish oils") OR TITLE-ABS("marine oil*") OR TITLE-ABS("algal oil*") OR TITLE-ABS(Fishes) OR TITLE-ABS(Fish) OR TITLE-ABS(seafood)

AND

TITLE-ABS-KEY ("Systematic review") OR TITLE-ABS-KEY ("Meta analysis") OR TITLE-ABS (review) OR TITLE-ABS (meta-analysis) OR TITLE-ABS (metaanalysis) OR TITLE-ABS ("quantitative review") OR TITLE-ABS ("quantitative overview") OR TITLE-ABS ("systematic overview") OR TITLE-ABS ("methodologic review") OR TITLE-ABS ("methodologic overview") OR TITLE-ABS("pooled analyses") OR TITLE-ABS("multi-center study") OR TITLE-ABS("multi-cohort study")

Limit: from 2000

A.2 Selection of systematic reviews including meta-analyses

Step 1. Identification
1739 records retrieved:
- PubMed: 676
- Scopus: 1063
- Other sources: 1

512 duplicates excluded

Step 2. Screening
1227 records screened,
1070 records excluded after first selection

Step 3. Eligibility
157 full-texts assessed,
130 records excluded after second selection due to:
- No exposure of interest: 9
- No outcome of interest: 4
- Different study design: 96
- Different study population: 6
- Updated version available: 7
- No subgroup analyses in CVD patients: 6
- Language: 1
- Different aim: 1

Step 4. Inclusion
25 records included for the evaluation of EPA and DHA (current background document)
2 records included for the evaluation of fish (different background document)

The selection procedure yielded 25 SRs that the Committee found suitable for the evaluation of EPA and DHA supplementation and another 2 SRs that were considered suitable for the evaluation of fish consumption. However, as explained in Chapter 2 of this background document, none of these 25 SRs/MAs as a whole were appropriate for the Committee's evaluation. Therefore, the Committee retrieved the relevant RCTs from the SRs/MAs. In total, 80 RCTs were identified from these SRs/MAs and were subsequently screened/assessed for eligibility, as described below.

Step 1. Identification
80 records (individual studies) retrieved from SRs/MAs

Step 2. Screening and Step 3. Eligibility
80 records screened/assessed,
61 records excluded due to:

- No exposure of interest: 15
- No outcome of interest: 8
- Different study design: 3
- Different study population: 15
- Study duration too short: 15
- Record used similar data as a record already included: 5

Step 4. Inclusion
19 records (RCTs) included

The Committee noted that another MA on the effect of EPA and DHA supplementation on health outcomes was published after the search data for this background document: Shen et al. (2022).[60] This MA does not contain any relevant studies other than those already selected by the Committee. It was therefore not included in this evaluation.

A.3 Search strategy for individual randomised controlled trials

The Committee performed a literature search to identify relevant individual randomised controlled trials that were not included in an MA. Publications were searched in PubMed and Scopus on 20th September 2021 using the following search strategies:

PubMed
(Coronary disease [MeSH] OR Acute coronary syndrome [MeSH] OR Angina pectoris [MeSH] OR Coronary artery disease [MeSH] OR Myocardial infarction [MeSH] OR Peripheral arterial disease [MeSH] OR Intermittent claudication [MeSH] OR Stroke [MeSH] OR Brain ischemia [MeSH] OR Cerebrovascular disorders [MeSH] OR Percutaneous coronary intervention [MeSH] OR Coronary artery bypass [MeSH] OR Coronary disease [TIAB] OR Coronary heart disease [TIAB] OR Acute coronary syndrome [TIAB] OR Angina pectoris [TIAB] OR Angina [TIAB] OR Ischemic heart disease [TIAB] OR Ischaemic heart disease [TIAB] OR Coronary artery disease [TIAB] OR Coronary Arteriosclerosis [TIAB] OR Myocardial infarction [TIAB] OR Heart attack [TIAB] OR Peripheral arterial disease [TIAB] OR Peripheral vascular disease [TIAB] OR Intermittent claudication [TIAB] OR Stroke [TIAB] OR Acute stroke [TIAB] OR Cerebrovascular Apoplexy [TIAB] OR Apoplexy [TIAB] OR Ischemic stroke [TIAB] OR Ischaemic stroke [TIAB] OR Hemorrhagic stroke [TIAB] OR Haemorrhagic stroke [TIAB] OR Cerebrovascular accident [TIAB] OR Acute cerebrovascular accident [TIAB] OR Cerebrovascular stroke [TIAB] OR Brain vascular accident [TIAB] OR Brain ischemia [TIAB] OR Cerebral ischemia [TIAB] OR Cerebral stroke [TIAB] OR Brain accident [TIAB] OR Brain infarction [TIAB] OR Cerebral infarction [TIAB] OR Transient

ischemic attack [TIAB] OR TIA [TIAB] OR Cerebrovascular* [TIAB] OR Subarachnoid haemorrhage [TIAB] OR Intracerebral hemorrhage [TIAB] OR Intracranial hemorrhages [TIAB] OR Coronary revascularization [TIAB] OR Percutaneous coronary intervention [TIAB] OR Coronary artery bypass graft surgery [TIAB] OR Percutaneous transluminal coronary angioplasty [TIAB] OR Percutaneous transluminal angioplasty [TIAB] OR Coronary angioplasty [TIAB] OR Atherosclerotic cardiovascular disease [TIAB] OR Carotid artery disease [TIAB] OR CHD [TIAB] OR ACS [TIAB] OR IHD [TIAB] OR CAD [TIAB] OR MI [TIAB] OR AMI [TIAB] OR PAD [TIAB] OR CVA [TIAB] OR CVAs [TIAB] OR TIA [TIAB] OR PCI [TIAB] OR CABG [TIAB] OR PTCA [TIAB] OR PTA [TIAB] OR ASCVD [TIAB])

AND

("Fatty Acids, Omega-3"[MeSH] OR omega-3 fatty acid* [TIAB] OR omega 3 fatty acid* [TIAB] OR n-3 fatty acid* [TIAB] OR n3 fatty acid* [TIAB] OR n 3 fatty acid* [TIAB] OR omega-3 [TIAB] OR omega 3 [TIAB] OR n-3 polyunsaturated [TIAB] OR n3 polyunsaturated [TIAB] OR n 3 polyunsaturated [TIAB] OR n-3 PUFA [TIAB] OR n3 PUFA [TIAB] OR Docosahexaenoic acid* [TIAB] OR Docosahexenoic acid* [TIAB] OR Eicosapentaenoic acid* [TIAB] OR Eicosapentenoic acid* [TIAB] OR DHA [TIAB] OR EPA [TIAB] OR fish fatty acid* [TIAB] OR Fish oils [MeSH] OR Fish oil* [TIAB] OR marine oil* [TIAB] OR algal oil* [TIAB])

AND

("Clinical Trials as Topic"[Mesh] OR "Clinical Trial" [publication type] OR "Cross-Over Studies"[Mesh] OR "Double-Blind Method"[Mesh] OR "Single-Blind Method"[Mesh] OR "Controlled Before-After Studies"[Mesh] OR "Historically Controlled Study"[Mesh] OR randomized[tiab] OR randomised[tiab] OR RCT[tiab] OR controlled*[tiab] OR placebo[tiab] OR clinical trial[tiab] OR trial[tiab] OR intervention[tiab])

NOT

("Systematic Review"[Publication Type] OR "Systematic Reviews as Topic"[MeSH Terms] OR "Review"[Publication Type] OR "meta analysis"[Publication Type] OR "Meta-Analysis as Topic"[MeSH Terms] OR "Network Meta-Analysis"[MeSH Terms] OR "Primary Prevention"[MeSH Terms])

Limit: from 2008

Scopus

TITLE-ABS("Coronary disease") OR TITLE-ABS("Acute coronary syndrome") OR TITLE-ABS("Angina pectoris") OR TITLE-ABS("Coronary artery disease") OR TITLE-ABS("Myocardial infarction") OR TITLE-ABS("Peripheral arterial disease") OR TITLE-ABS("Intermittent claudication") OR TITLE-ABS(Stroke) OR TITLE-ABS("Brain ischemia") OR TITLE-ABS("Cerebrovascular disorders") OR TITLE-ABS("Percutaneous coronary intervention") OR TITLE-ABS("Coronary artery bypass") OR TITLE-ABS("Coronary heart disease") OR TITLE-ABS(Angina) OR TITLE-ABS("Ischemic heart disease") OR TITLE-ABS("Ischaemic heart disease") OR TITLE-ABS("Coronary Arteriosclerosis") OR TITLE-ABS("Heart attack") OR TITLE-ABS("Peripheral vascular disease") OR TITLE-ABS("Acute stroke") OR TITLE-ABS("Cerebrovascular Apoplexy") OR TITLE-ABS(Apoplexy) OR TITLE-ABS("Ischemic stroke") OR TITLE-ABS("Ischaemic stroke") OR TITLE-ABS("Hemorrhagic stroke") OR TITLE-ABS("Haemorrhagic stroke") OR TITLE-ABS("Cerebrovascular accident") OR TITLE-ABS("Acute cerebrovascular accident") OR TITLE-ABS("Cerebrovascular stroke") OR TITLE-ABS("Brain vascular accident") OR TITLE-ABS("Brain ischemia") OR TITLE-ABS("Cerebral ischemia") OR TITLE-ABS("Cerebral stroke") OR TITLE-ABS("Brain accident") OR TITLE-ABS("Brain infarction") OR TITLE-ABS("Cerebral infarction") OR TITLE-ABS("Transient ischemic attack") OR TITLE-ABS(TIA) OR TITLE-ABS(Cerebrovascular*) OR TITLE-ABS("Subarachnoid haemorrhage") OR TITLE-ABS("Intracerebral hemorrhage") OR TITLE-ABS("Intracranial hemorrhages") OR TITLE-ABS("Coronary revascularization") OR TITLE-ABS("Percutaneous coronary intervention") OR TITLE-ABS("Coronary artery bypass graft surgery") OR TITLE-ABS("Percutaneous transluminal coronary angioplasty") OR TITLE-ABS("Percutaneous transluminal angioplasty") OR TITLE-ABS("Coronary angioplasty") OR TITLE-ABS("Atherosclerotic cardiovascular disease") OR TITLE-ABS("Carotid artery disease") OR TITLE-ABS(CHD) OR TITLE-ABS(ACS) OR TITLE-ABS(IHD) OR TITLE-ABS(CAD) OR TITLE-ABS(MI) OR TITLE-ABS(AMI) OR TITLE-ABS(PAD) OR TITLE-ABS(CVA) OR TITLE-ABS(CVAs) OR TITLE-ABS(TIA) OR TITLE-ABS(PCI) OR TITLE-ABS(CABG) OR TITLE-ABS(PTCA) OR TITLE-ABS(PTA) OR TITLE-ABS(ASCVD)

AND

TITLE-ABS("Fatty Acids, Omega-3") OR TITLE-ABS("omega 3 fatty acid*") OR TITLE-ABS("n-3 fatty acid*") OR TITLE-ABS("n3 fatty acid*") OR ("n 3 fatty acid*") OR TITLE-ABS(omega-3) OR TITLE-ABS("omega 3") OR TITLE-ABS("n-3 polyunsaturated") OR TITLE-ABS("n3 polyunsaturated") OR TITLE-ABS("n 3 polyunsaturated") OR TITLE-ABS("n-3 PUFA") OR TITLE-ABS("n3 PUFA") OR TITLE-ABS("Docosahexaenoic acid*") OR TITLE-ABS("Docosahexenoic acid*") OR TITLE-ABS("Eicosapentaenoic acid*") OR TITLE-ABS("Eicosapentenoic acid*") OR TITLE-ABS(DHA) OR TITLE-

ABS(EPA) OR TITLE-ABS("fish fatty acid*") OR TITLE-ABS("Fish oils") OR TITLE-ABS("marine oil*") OR TITLE-ABS("algal oil*")

AND

TITLE-ABS-KEY ("Clinical Trial") OR TITLE-ABS-KEY ("Cross-Over Studies") OR TITLE-ABS-KEY("Double-Blind Method") OR TITLE-ABS-KEY("Single-Blind Method") OR TITLE-ABS-KEY("Controlled Before-After Studies") OR TITLE-ABS-KEY("Historically Controlled Study") OR TITLE-ABS-KEY(randomized) OR TITLE-ABS-KEY(randomised) OR TITLE-ABS-KEY(RCT) OR TITLE-ABS-KEY(controlled*) OR TITLE-ABS-KEY(placebo) OR TITLE-ABS-KEY("clinical trial") OR TITLE-ABS-KEY(trial) OR TITLE-ABS-KEY(intervention)

AND NOT

TITLE-ABS-KEY ("Systematic Review") OR TITLE-ABS-KEY (Review) OR TITLE-ABS-KEY ("Meta-Analysis") OR TITLE-ABS-KEY ("Meta Analysis") OR TITLE-ABS-KEY ("Network Meta-Analysis") OR TITLE-ABS-KEY ("Primary Prevention")

Limit: from 2008

A.4 Selection of individual randomised controlled trials

Step 1. Identification
487 records retrieved:
- PubMed: 152
- Scopus: 332
- Other sources: 3

100 duplicates excluded

Step 2. Screening
387 records screened,
369 records excluded after first selection

Step 3. Eligibility
18 full-texts assessed,
10 records excluded after second selection due to:
- No exposure of interest: 2
- No outcome of interest: 2
- Insufficient analyses: 2
- Different study population: 2

- Record used similar data as a record already included: 2

Step 4. Inclusion
8 records included

This search for individual RCTs yielded 8 publications that the Committee considered suitable for the evaluation of EPA and DHA supplementation. Together with the 19 publications retrieved via SRs/MAs (see above in section A.2), a total of 27 publications were included in the Committee's evaluation of EPA and DHA supplementation.

Annex B Overview of selected RCTs

Table B1 Overview of selected RCTs on the effect of EPA and DHA supplementation on health outcomes in people with ASCVD, according to the EPA/DHA dose (≤1 g/d and >1 g/d)[a]

Health outcome	RCTs with an EPA/DHA dose of ≤1 g/d	RCTs with an EPA/DHA dose of >1 g/d
All-cause mortality	Alpha Omega[28] GISSI-P[30] Nutristroke[31] OMEGA[32] SU.FOL.OM3[34]	HARP[38] HEARTS[39] IEIS-4[26] NAT2[42] Nosaka et al., 2017[21] OFAMI[43] OMEGA-REMODEL[44] OMEMI[45] OPACH[46] SCIMO[48] SHOT[49] STRENGTH[50]
Total CVD/MACE	Alpha Omega[28] GISSI-P[30] Nutristroke[31] OMEGA[32] SU.FOL.OM3[34]	HARP[38] HEARTS[39] IEIS-4[26] JELIS[23] Nosaka et al., 2017[21] OFAMI[43] OMEMI[45] OPACH[46] REDUCE-IT[20] SCIMO[48] SHOT[49] STRENGTH[50]
Fatal CVD	Alpha Omega[28] GISSI-P[30] Nutristroke[31] OMEGA[32] SU.FOL.OM3[34]	HARP[38] Nosaka et al., 2017[21] OFAMI[43] SCIMO[48] SHOT[49] STRENGTH[50]
Non-fatal CVD	GISSI-P[30]	IEIS-4[26] OFAMI[43] SCIMO[48]
Total CHD	Alpha Omega[28] GISSI-P[30] OMEGA[32] SU.FOL.OM3[34]	HEARTS[39] IEIS-4[26] JELIS[23] Nye et al., 1990[22] OFAMI[43] OPACH[46]

		SCIMO[48] SHOT[49] STRENGTH[50]
Fatal CHD	Alpha Omega[28] GISSI-P[30] OMEGA[32] SU.FOL.OM3[34]	HARP[38] IEIS-4[26] JELIS[23] OFAMI[43] SCIMO[48] SHOT[49]
Non-fatal CHD	N/A	JELIS[23] OFAMI[43]
Total MI	Alpha Omega[28] GISSI-P[30] OMEGA[32] SU.FOL.OM3[34]	HARP[38] JELIS[23] Nosaka et al., 2017[21] OFAMI[43] OPACH[46] SCIMO[48] SHOT[49]
Fatal MI	Alpha Omega[28] GISSI-P[30,36] OMEGA[32] SU.FOL.OM3[34]	HARP[38] IEIS-4[26] JELIS[23] SCIMO[48] SHOT[49]
Non-fatal MI	Alpha Omega[28] GISSI-P[30] OMEGA[32] SU.FOL.OM3[34]	HARP[38] IEIS-4[26] JELIS[23] Nosaka et al., 2017[21] OFAMI[43] OMEMI[45] SCIMO[48] SHOT[49]
Total stroke	Alpha Omega[28] GISSI-P[30] OMEGA[32] SU.FOL.OM3[34]	HARP[38] JELIS[40] NAT2[42] Nosaka et al., 2017[21] OFAMI[43] OMEMI[45] OPACH[46] SCIMO[48]
Non-fatal stroke	N/A	Nosaka et al., 2017[21]
Sudden death	GISSI-P[30] OMEGA[32] SU.FOL.OM3[34]	IEIS-4[26] JELIS[23] Nosaka et al., 2017[21] OFAMI[43] SHOT[49]

Revascularisation	GISSI-P[36] OMEGA[32] SU.FOL.OM3[34]	HARP[38] JELIS[23] Nosaka et al., 2017[21] OFAMI[43] OMEMI[45] OPACH[46] REDUCE-IT[47] SCIMO[48]
Angina pectoris	GISSI-P[36] OMEGA[32]	HARP[38] IEIS-4[26] JELIS[23] Nye et al., 1990[22] OFAMI[43] SCIMO[48]
Heart failure	OMEGA[32] SU.FOL.OM3[34]	HARP[38] Nosaka et al., 2017[21] OMEMI[45]
Arrhythmia	Alpha Omega[28] GISSI-P[30] OMEGA[32] SU.FOL.OM3[34]	OFAMI[43] OMEMI[45]
PAD progression	ORIGIN[33]	N/A
Depression	Alpha Omega[29] SU.FOL.OM3[35]	N/A
Cancer	GISSI-P[36] OMEGA[32] SU.FOL.OM3[34]	N/A
Systolic blood pressure	Alpha Omega[28] SU.FOL.OM3[34]	Gans et al., 1990[37] HARP[38] HEARTS[39] JELIS[23] OFAMI[43] SCIMO[48]
LDL cholesterol	Alpha Omega[28]	Gans et al., 1990[37] HARP[38] HEARTS[39] JELIS[23] Mori et al., 1992[41] NAT2[42] SCIMO[48] SHOT[49]
Body weight	N/A	HARP[38] HEARTS[39] SCIMO[48]

Abbreviations: ASCVD: atherosclerotic cardiovascular disease; CHD: coronary heart disease; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; LDL: low-density lipoprotein, MACE: major adverse cardiovascular events, MI: myocardial infarction; N/A: not applicable; PAD: peripheral arterial disease; RCT: randomised controlled trial.

Footnotes:

[a] Health outcomes for which no literature was found were not reported in the table.

Annex C Description and results of selected RCTs

Table C1 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Alfaddagh et al. (HEARTS) and Bhatt et al. and Peterson et al. (REDUCE-IT)

Aspect	Alfaddagh et al. 2017[39]	Bhatt et al. 2019[20] and Peterson et al. 2021[47]
Trial name	HEARTS	REDUCE-IT
Study duration	30 months[a]	Median 5 years (maximum 6.2 y)
Primary disease	CHD	CHD, stroke, PAD
Study design	Parallel RCT (open-label)	Parallel RCT
Number of participants; health outcome; number of cases	Participants: i: 143, c: 142; ALL-CAUSE MORTALITY: Cases: i: 1, c: 3 TOTAL CVD: Cases: i: 45, c: 39 TOTAL CHD: Cases: i: 41, c: 35 SYSTOLIC BLOOD PRESSURE and LDL CHOLESTEROL and BODY WEIGHT: Participants: i: 126, c: 114	Participants: i: 2892, c: 2893; TOTAL CVD: Cases: i: 559, c: 738 REVASCULARISATION: Cases: i: 306, c: 464
Diet of intervention (i) and control (c) group	i: 3.36 g/d long-chain n-3 ethyl esters from fish oil (Lovaza®) (1.86 g/d EPA, 1.5 g/d DHA) c: nil (no placebo)	i: 4 g/d of EPA ethyl ester supplements (icosapent ethyl; VASCEPA®) c: mineral oil (paraffin oil)
Strength of the effect: HR or RR (95%CI) or MD (95%CI)	ALL-CAUSE MORTALITY: 0.33 (0.03, 3.14) TOTAL CVD: 1.15 (0.80, 1.64) TOTAL CHD: 1.16 (0.79, 1.71) SYSTOLIC BLOOD PRESSURE: Between-group MD: -0.3 (-4.43, 3.83) mmHg LDL CHOLESTEROL: Between-group MD: 0.08 (-0.11, 0.27) mmol/L	TOTAL CVD: 0.73 (0.65, 0.81) REVASCULARISATION: 0.63 (0.55, 0.73)
Study population	People with stable CHD on statins; BMI: NR; medication in c group: statins, aspirin, ACE inhibitors, and β-blockers (>50%), ARB, hydrochlorothiazide, and calcium channel blocker (20-49%); men (85%) and women; USA	People with documented CHD, cerebrovascular or carotid disease or documented PAD, and with a fasting triglyceride level of 150-499 mg/dL and a LDL cholesterol level of 41-100 mg/dL, and on stable statin therapy; BMI[b]: 31 (28-35) kg/m^2;

		medication: statins (100%); men (~71%) and women; North-America, Europe, Australia, South Africa, Asia
Compliance	Unclear	NR

Abbreviations: ACE: angiotensin converting enzyme; ARB: angiotensin receptor blocker; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; HEARTS: Slowing HEART diSease with lifestyle and omega-3 fatty acids; HR: hazard ratio; i: intervention group; IQR: interquartile range; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; NR: not reported; PAD: peripheral arterial disease; RCT: randomised controlled trial; REDUCE-IT: Reduction of Cardiovascular Events with Icosapent Ethyl-Intervention Trial; RR: relative risk; USA: United States of America.

Footnotes:

[a] Refers to the duration of the intervention.

[b] Median (± interquartile range) for the total cohort, i.e. the secondary prevention cohort (as described in the current table; ~71% of the total cohort) and the primary-prevention cohort (people with a high risk of CVD; ~29% of the total cohort) combined. Median/mean BMI was not reported for the primary-prevention cohort and the secondary-prevention cohort separately.

Table C2 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Dagenais et al. (ORIGIN) and Eritsland et al. (SHOT)

Aspect	Dagenais et al. 2019[33]	Eritsland et al. 1996[49]
Trial name	ORIGIN	SHOT
Study duration	6.2 years (median)	12 months (maximum)
Primary disease	PAD	CHD
Study design	Parallel, 2x2 factorial RCT	Parallel, 2x2 factorial RCT (open-label)
Number of participants; health outcome; number of cases	Participants: i: 485, c: 486; PAD PROGRESSION[a]: Cases: i: 70, c: 76	Participants: i: 317, c: 293; ALL-CAUSE MORTALITY: Cases: i: 8, c: 6 TOTAL CVD: Cases: i: 15, c: 12 FATAL CVD: Cases: i: 7, c: 5 TOTAL CHD: Cases: i: 7, c: 12 FATAL CHD: Cases: i: 7, c: 4 TOTAL MI: Cases: i: 7, c: 12 FATAL MI: Cases: i: 7, c: 4 NON-FATAL MI: Cases: i: 5, c: 3 SUDDEN DEATH:

		Cases: i: 7, c: 4 LDL CHOLESTEROL: Participants: i: 289, c: 267
Diet of intervention (i) and control (c) group	i: 1 g/d omega-3 acid ethyl esters (Omacor®; 465 mg EPA + 375 mg DHA) c: 1 g/d olive oil	i: 3.3 g/d of n-3 PUFA (Omacor®; 2.04 g EPA + 1.28 g DHA) c: nil (no placebo)
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	PAD PROGRESSION: 0.90 (0.65, 1.24)	ALL-CAUSE MORTALITY: 1.23 (0.43, 3.51) TOTAL CVD: 1.16 (0.55, 2.43) FATAL CVD: 1.29 (0.42, 4.03) TOTAL CHD: 0.54 (0.22, 1.35) FATAL CHD: 1.62 (0.48, 5.47) TOTAL MI: 0.54 (0.22, 1.35) FATAL MI: 1.62 (0.48, 5.47) NON-FATAL MI: 1.54 (0.37, 6.39) SUDDEN DEATH: 1.62 (0.48, 5.47) LDL CHOLESTEROL: Between-group MD: 0.08 (-0.12, 0.28) mmol/L
Study population	People with PAD (ABI ≤0.9) and diabetes or prediabetes; BMI: 29 ± 5 kg/m^2; medication: statins (53%), β-blockers (48%), ACE inhibitors or ARB (72%), diuretics (36%), aspirin or antiplatelets (68%), metformin (23%); men (~66%) and women; Europe and USA	People admitted for CABG; BMI[b]: ~25 (±3) kg/m^2; medication: NR; men (~87%) and women; Europe
Compliance	Adherence to the intervention was 83% and did not differ between subgroups at the end of follow-up.	88% of the fish oil capsules were taken

Abbreviations: ABI: ankle-brachial index; ACE: angiotensin converting enzyme; ARB: angiotensin receptor blocker; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CABG: coronary artery bypass graft; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; NR: not reported; ORIGIN: Outcome Reduction with Initial Glargine Intervention; PAD: peripheral arterial disease; PUFA: polyunsaturated fatty acid; RCT: randomised controlled trial; RR: relative risk; SHOT: Shunt Occlusion Trial; USA: United States of America.

Footnotes:

[a] Dagenais et al. defined PAD progression as the occurrence of either a decrease of at least 0.1 unit in ABI as compared to baseline ABI (asymptomatic PAD) or claudication with an ABI of 0.90 or less or lower limb revascularisation or amputation as the result of arterial disease (symptomatic PAD).

[b] Mean (± standard deviation).

Table C3 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Galan et al. and Andreeva et al. (SU.FOL.OM3) and Gans et al.

Aspect	Galan et al. 2010[34] and Andreeva et al. 2012[35]	Gans et al. 1990[37]
Trial name	SU.FOL.OM3	N/A
Study duration	4 years[a]	4 months
Primary disease	CHD, stroke	PAD
Study design	Parallel, 2x2 factorial RCT	Parallel RCT
Number of participants; health outcome; number of cases	Participants: i: 1253, c: 1248; ALL-CAUSE MORTALITY: Cases: i: 58, c: 59 TOTAL CVD: Cases: i: 216, c: 211 FATAL CVD: Cases: i: 23, c: 28 TOTAL CHD: Cases: i: 37, c: 41 FATAL CHD: Cases: i: 5, c: 13 TOTAL MI: Cases: i: 32, c: 32 FATAL MI: Cases: i: 1, c: 2 NON-FATAL MI: Cases: i: 32, c: 28 SUDDEN DEATH: Cases: i: 4, c: 11 REVASCULARISATION: Cases: i: 152, c: 156 HEART FAILURE: Cases: i: 21, c: 22 ARRHYTHMIA: Cases: i: 33, c: 32 CANCER: Cases: i: 88, c: 81 DEPRESSIVE SYMPTOMS[b]: Cases: 134 (total) SYSTOLIC BLOOD PRESSURE: Participants: i: 1253, c: 1248	Participants: i: 16, c: 17

Diet of intervention (i) and control (c) group	i: omega-3 supplements providing 400 mg/d EPA and 200 mg/d DHA c: gelatin capsules (placebo)	i: 1.8 g/d of EPA and 1.8 g/d of DHA (fish oil) c: corn oil
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	ALL-CAUSE MORTALITY: 0.98 (0.69, 1.39) TOTAL CVD: 1.02 (0.86, 1.21) FATAL CVD: 0.82 (0.47, 1.41) TOTAL CHD: 0.90 (0.58, 1.39) FATAL CHD: 0.38 (0.14, 1.07) TOTAL MI: 1.00 (0.61, 1.62) FATAL MI: 0.5 (0.05, 5.49) NON-FATAL MI: 1.14 (0.69, 1.88) SUDDEN DEATH: 0.36 (0.12, 1.13) REVASCULARISATION: 0.97 (0.79, 1.20) HEART FAILURE: 0.95 (0.53, 1.72) ARRHYTHMIA: 1.03 (0.64, 1.66) CANCER: 1.10 (0.81, 1.48) DEPRESSIVE SYMPTOMS: 1.16 (0.95, 1.41) SYSTOLIC BLOOD PRESSURE: Between-group mean difference: -0.06 mmHg (-0.90, 0.80)	SYSTOLIC BLOOD PRESSURE: Between-group MD: 5 (-11.59, 21.59) mmHg LDL CHOLESTEROL: Between-group MD: 0.62 (-0.01, 1.25) mmol/L
Study population	People with a history of MI, unstable angina or ischaemic stroke; BMI: NR; medication: lipid-lowering drugs (86%), β-blockers, aspirin or antiplatelets and ACE inhibitors (>50%); men (~80%) and women; Europe	People with symptoms of intermittent claudication due to atherosclerotic disease; BMI: NR; men (69%); Europe
Compliance	Compliance: 86% (defined as taking at least 80% of treatment). Compliance was self-reported (questionnaires)	NR

Abbreviations: ACE: angiotensin converting enzyme; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; N/A: not applicable; NR: not reported; PAD: peripheral arterial disease; RCT: randomised controlled trial; RR: relative risk; SU.FOL.OM3: Supplémentation en Folates et omega-3.

Footnotes:

[a] Refers to the duration of the intervention.

[b] Depressive symptoms were assessed using the 30-item Geriatric Depression Scale (GDS). A dichotomous outcome was defined: GDS ≤10 (no depressive symptoms) versus GDS >10 (any level of depressive symptoms).

Table C4 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Garbagnati et al. (Nutristroke) and Heydari et al. (OMEGA-REMODEL)

Aspect	Garbagnati et al. 2009[31]	Heydari et al. 2016[44]
Trial name	Nutristroke	OMEGA-REMODEL
Study duration	12 months[a]	24 months (median)
Primary disease	Stroke	CHD
Study design	Parallel RCT	Parallel RCT
Number of participants; health outcome; number of cases	Participants: i: 38, c: 34; ALL-CAUSE MORTALITY: Cases: i: 0, c: 3 TOTAL CVD: Cases: i: 0, c: 4 FATAL CVD: Cases: i; 0, c: 4	Participants: i: 180, c: 178; ALL-CAUSE MORTALITY: Cases: i: 8, c: 2
Diet of intervention (i) and control (c) group	i: 0.5 g/d EPA and DHA, supplement containing 250 mg EPA and 250 mg DHA c: unclear placebo (identical to supplement but contained no antioxidants or PUFAs)	i: 4 g/d of EPA/DHA ethyl ester supplements (Lovaza®) containing 1.9 g/d EPA and 1.5 g/d DHA c: corn oil placebo supplements (placebo) containing 3.2 g/d LA and no EPA or DHA
Strength of the effect: HR or RR (95%CI)	ALL-CAUSE MORTALITY: 0.13 (0.01, 2.34) TOTAL CVD: 0.10 (0.01, 1.79) FATAL CVD: 0.10 (0.01, 1.79)	ALL-CAUSE MORTALITY: 3.96 (0.85, 18.37)
Study population	People in a rehabilitation unit who have survived a stroke; BMI: NR; medication: NR; men (i: 74%, c: 56%) and women; Europe	People with acute MI; BMI[b]: 29± 6 kg/m^2; medication: dual antiplatelet (98%), beta-blockers (92%), statins (97%), calcium-channel blockers (7%), ACE inhibitors or ARB (73%), diuretics (13%); men (65%) and women; USA

Compliance	Assessed but unclear results	96% in both i and c group (*P*=0.86) (assessed by capsule count)

Abbreviations: ACE: angiotensin converting enzyme; ARB: angiotensin receptor blocker; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; LA: linoleic acid; MI: myocardial infarction; NR: not reported; PUFA: polyunsaturated fatty acid; RCT: randomised controlled trial; RR: relative risk; USA: United States of America.

Footnotes:

[a] Refers to the duration of the intervention.

[b] Mean (± standard deviation).

Table C5 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Kalstad et al. (OMEMI) and Kromhout et al. and Giltay et al. (Alpha Omega)

Aspect	Kalstad et al. 2021[45]	Kromhout et al. 2010[28] and Giltay et al. 2011[29]
Trial name	OMEMI	Alpha Omega
Study duration	2 years (maximum)	40 months[a]
Primary disease	CHD	CHD
Study design	Parallel RCT	Parallel, 2x2 factorial RCT
Number of participants; health outcome; number of cases	Participants; i: 505, c: 509 ALL-CAUSE MORTALITY: Cases: i: 28, c: 28 TOTAL CVD: Cases: i: 108, c: 102 NON-FATAL MI: Cases: i: 39, c: 35 TOTAL STROKE: Cases: i: 17, c: 12 REVASCULARISATION: Cases: i: 14, c: 21 HEART FAILURE: Cases: i: 20 c: 17 ATRIAL FIBRILLATION: Cases: i: 28, c: 15	Participants: i: 2404, c: 2433; ALL-CAUSE MORTALITY: Cases: i: 186, c: 184 TOTAL CVD: Cases: i: 332, c: 331 FATAL CVD: Cases: i: 80, c: 82 TOTAL CHD: Cases: i: 120, c: 128 FATAL CHD: Cases: i: 67, c: 71 TOTAL MI: Cases: i: 89, c: 102 FATAL MI: Cases: i: 36, c: 102 NON-FATAL MI: Cases: i: 56, c: 59 TOTAL STROKE: Cases: i: 11, c: 10 ARRHYTHMIA: Cases: i: 67, c: 74 SEVERE DEPRESSIVE SYMPTOMS[b]: Cases: i: 15, c: 13 SYSTOLIC BLOOD PRESSURE: Participants: i: 1192, c: 1236

		LDL CHOLESTEROL: Participants: i: 562, c: 562
Diet of intervention (i) and control (c) group	i: 1.8 g/d n-3 PUFA (Pikasol®; 930 mg EPA + 660 mg DHA) c: corn oil (placebo)	i: 20 g/d margarine enriched with 240 mg EPA and 160 mg DHA (or 20 g/d margarine enriched with 240 mg EPA, 150 mg DHA and 2 g/d ALA) c: 20 g/d margarine (placebo) (or 20 g/d margarine enriched with 2 g/d ALA)
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	ALL-CAUSE MORTALITY: 1.01 (0.60, 1.71) TOTAL CVD: 1.07 (0.82, 1.40) NON-FATAL MI: 1.14 (0.72, 1.80) TOTAL STROKE: 1.37 (0.65, 2.88) REVASCULARISATION: 0.66 (0.34, 1.30) HEART FAILURE: 1.19 (0.62, 2.26) ATRIAL FIBRILLATION: 1.84 (0.98, 3.45)	ALL-CAUSE MORTALITY: 1.02 (0.84, 1.24) TOTAL CVD: 1.02 (0.88, 1.17) FATAL CVD: 0.99 (0.73, 1.34) TOTAL CHD: 0.95 (0.74, 1.21) FATAL CHD: 0.95 (0.68, 1.32) TOTAL MI: 0.88 (0.67, 1.17) FATAL MI: 0.36 (0.25, 0.52) NON-FATAL MI: 0.96 (0.67, 1.38) TOTAL STROKE: 1.11 (0.47, 2.62) ARRHYTHMIA: 0.92 (0.66, 1.27) SEVERE DEPRESSIVE SYMPTOMS: 1.29 (0.60, 2.78) SYSTOLIC BLOOD PRESSURE: Between-group MD: 1.70 (-0.60, 3.90) mmHg LDL CHOLESTEROL: Between-group MD: -0.02 (-0.1, 0.06) mmol/L
Study population	People who had an acute MI; BMI[c]: 27 kg/m^2; medication: aspirin (94%), other antiplatelet therapy (89%), anticoagulants (18%), statins (97%), antihypertensives (72%), β-blockers (83%); men (71%) and women; Europe (Norway)	People with previous MI; BMI: NR; medication: lipid-lowering drugs (85%), antihypertensives (90%), antithrombotics (98%), antiarrhythmic drugs (3%); men (78%) and women; Europe

Compliance	Self-reported adherence in 88% of participants	90.5% compliance and consumed 20.6 (±2.8) g/d of margarine (assessed by measuring daily intakes of margarine and n-3 FAs through returned margarine tubs. Adherence measured by levels of FAs in plasma cholesteryl esters, margarine and questionnaires)

Abbreviations: ALA: alpha linoleic acid; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; FA: fatty acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; NR: not reported; OMEMI: Omega-3 Fatty acids in Elderly with Myocardial Infarction; RCT: randomised controlled trial; RR: relative risk.
Footnotes:
[a] Refers to the duration of the intervention.
[b] Depressive symptoms were assessed using the 15-item Geriatric Depression Scale (GDS). A dichotomous outcome was defined: GDS ≤10 (no severe depressive symptoms) versus GDS >10 (severe depressive symptoms).
[c] Mean (± standard deviation).

Table C6 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Marchioli et al. (GISSI-P) and Mori et al.

Aspect	Marchioli et al. 1999[30]	Mori et al. 1992[41]
Trial name	GISSI-P	N/A
Study duration	42 months (maximum)	10 weeks
Primary disease	CHD	PAD
Study design	Parallel, 2x2 factorial RCT (open-label)	RCT
Number of participants; health outcome; number of cases	Participants: i: 5666, c: 5658; ALL-CAUSE MORTALITY: Cases: i: 472, c: 545 TOTAL CVD: Cases: i: 1552, c: 1550 FATAL CVD: Cases: i: 291, c: 348 NON-FATAL CVD: Cases: i: 287, c: 291 TOTAL CHD: Cases: i: 424, c: 485 FATAL CHD: Cases: i: 247, c: 306 TOTAL MI: Cases: i: 223, c: 233 FATAL MI: Cases: i: 214, c: 265 NON-FATAL MI: Cases: i: 223, c: 233	Participants: i: 15, c: 14

	TOTAL STROKE: Cases: i: 98, c: 80 SUDDEN DEATH: Cases: i: 122, c: 164 REVASCULARISATION: Cases: i: 1104, c: 1053 ANGINA PECTORIS: Cases: i: 254, c: 249 ARRHYTHMIA: Cases: i: 40, c: 46 CANCER: Cases: i: 77, c: 61	
Diet of intervention (i) and control (c) group	i: 1 g/d omega-3 acid ethyl esters (Omacor®; 880 mg EPA+DHA) c: nil (no placebo)	i: 2.8 g/d of EPA and 1.8 g/d of DHA (fish oil) c: olive oil
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	ALL-CAUSE MORTALITY: 0.86 (0.77, 0.97) TOTAL CVD: 1.00 (0.94, 1.06) FATAL CVD: 0.84 (0.72, 0.97) NON-FATAL CVD: 0.99 (0.83, 1.17) TOTAL CHD: 0.87 (0.77, 0.99) FATAL CHD: 0.78 (0.66, 0.92) TOTAL MI: 0.96 (0.80, 1.14) FATAL MI: 0.80 (0.70, 0.90) NON-FATAL MI: 0.96 (0.80, 1.14) TOTAL STROKE: 1.22 (0.91, 1.64) SUDDEN DEATH: 0.74 (0.58, 0.93) REVASCULARISATION: 1.05 (0.98, 1.13) ANGINA PECTORIS: 1.02 (0.86, 1.20) ARRHYTHMIA: 0.87 (0.57, 1.32) CANCER: 1.26 (0.99, 1.77)	LDL CHOLESTEROL: Between-group mean difference: 1.01 (0.33, 1.69) mmol/L

Study population	People with recent MI; BMI: NR; medication: anti-platelets (90%), ACE inhibitors (47%), β-blockers (44%), lipid-lowering medication (baseline: 5%, after 42 months: 46%); men (~85%) and women; Europe	Participants with PAD; BMI: 25 (±1) kg/m^2; medication: antihypertensives (53%), hypolipidemic agents (3%); men (100%); Australia
Compliance	11.6% stopped taking Omacor by 12 mo, 28.5% by the end of the trial (assessed by capsule counts)	Compliance, as determined by capsule count, was 99% and 98% in the intervention group and control group, respectively.

Abbreviations: ACE: angiotensin converting enzyme; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; GISSI-P: Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardio (GISSI)-Prevenzione; HR: hazard ratio; i: intervention group; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; N/A: not applicable; NR: not reported; PAD: peripheral arterial disease; RCT: randomised controlled trial; RR: relative risk.

Table C7 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Nicholls et al. (STRENGTH) and Nilsen et al. (OFAMI)

Aspect	Nicholls et al. 2020[50]	Nilsen et al. 2001[43]
Trial name	STRENGTH	OFAMI
Study duration	42 months (median)	1.5 years (median)
Primary disease	TOTAL CVD	CHD
Study design	Parallel RCT	Parallel RCT
Number of participants; health outcome; number of cases	Participants: i: 3638, c: 3678; ALL-CAUSE MORTALITY: Cases: i: 234, c: 202 TOTAL CVD: Cases: i: 569, c: 610 FATAL CVD: Cases: i: 152, c: 138 TOTAL CHD: Cases: i: 417, c: 493	Participants: i: 150, c: 150; ALL-CAUSE MORTALITY: Cases: i: 11, c: 11 TOTAL CVD: Cases: i: 42, c: 36 FATAL CVD: Cases: i: 8, c: 8 NON-FATAL CVD: Cases: i: 39, c: 31 TOTAL CHD: Cases: i: 42, c: 36 FATAL CHD: Cases: i: 8, c; 8 NON-FATAL CHD: Cases: i: 39, c: 31 TOTAL MI: Cases: i: 21, c: 15 NON-FATAL MI: Cases: i: 21, c: 15

		TOTAL STROKE: Cases: i: 6, c: 0 SUDDEN DEATH: Cases: i: 0, c: 1 REVASCULARISATION: Cases: i: 43, c: 49 ANGINA PECTORIS” Cases: i: 26, c: 23 ARRHYTHMIA: Cases: i: 8, c: 15 SYSTOLIC BLOOD PRESSURE: Participants: i: 127, c: 130
Diet of intervention (i) and control (c) group	i: 4 g/d omega 3 carboxylic acid (EPA+DHA; Epanova®) c: corn oil	i: 4 g/d EPA/DHA ethyl ester supplements (Omacor®) containing 3.46 g/d EPA+DHA c: corn oil supplements (4 g/d)
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	ALL-CAUSE MORTALITY: 1.18 (0.97, 1.42) TOTAL CVD: 0.94 (0.84, 1.05) FATAL CVD: 1.12 (0.89, 1.41) TOTAL CHD: 0.85 (0.75, 0.97)	ALL-CAUSE MORTALITY: 1.00 (0.45, 2.24) TOTAL CVD: 1.17 (0.80, 1.71) FATAL CVD: 1.00 (0.39, 2.59) NON-FATAL CVD: 1.35 (0.79, 2.31) TOTAL CHD: 1.17 (0.80, 1.71) FATAL CHD: 1.00 (0.39, 2.59) NON-FATAL CHD: 1.30 (0.81, 2.08) TOTAL MI: 1.40 (0.75, 2.61) NON-FATAL MI: 1.40 (0.75, 2.61) TOTAL STROKE: 13 (0.74, 228.73) SUDDEN DEATH: 0.33 (0.01, 8.19) REVASCULARISATION: 0.83 (0.51, 1.35) ANGINA PECTORIS: 1.16 (0.63, 2.14) ARRHYTHMIA: 0.53 (0.23, 1.22) SYSTOLIC BLOOD PRESSURE: -3.80 (-9.19, 1.59)

Study population	People with established atherosclerotic CVD involving the coronary, peripheral, carotid, or aortic territories; BMI: NR; medication: statins (100%), RAAS blockers (% NR), antiplatelet agents (% NR), β-blockers (% NR); men (~65%) and women; North America, Europe, South America, Australia, Asia, New Zealand, and South Africa	People with acute MI; BMI[a]: 26 (17-42) kg/m^2; medication[b]: β-blockers(~56%), ACE inhibitors~20%), diuretics (~20%), aspirin (~865), statins (at hospital discharge: i: 6%, c: 8%; after 12 months: i: 71%, c: 65%); men (~80%) and women; Europe
Compliance	NR	i: 82% and c: 86% after 6 weeks (assessed by questionnaire and capsule count)

Abbreviations: ACE: angiotensin converting enzyme; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; MD: mean difference; MI: myocardial infarction; N/A: not applicable; NR: not reported; OFAMI: Omacor Following Acute Myocardial Infarction; RAAS: renin-angiotensin-aldosterone system; RCT: randomised controlled trial; RR: relative risk; STRENGTH: Long-Term Outcomes Study to Assess Statin Residual Risk with Epanova in High Cardiovascular Risk Patients with Hypertriglyceridemia.

Footnotes:

[a] Mean (± standard deviation).

[b] Represents medication use after 12 months. Additional data available for medication before admission, at the first 24 hours and at discharge (but not reported in the table).

Table C8 Summary of study characteristics and effects of EPA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Nosaka et al. and Nye et al.

Aspect	Nosaka et al. 2017[21]	Nye et al. 1990[22]
Trial name	N/A	N/A
Study duration	1 year[a]	12 months[a]
Primary disease	CHD	CHD
Study design	Parallel RCT (open-label)	Parallel RCT
Number of participants; health outcome; number of cases	Participants: i: 119, c: 119; ALL-CAUSE MORTALITY: Cases: i: 2, c: 9 TOTAL CVD: Cases: i: 11, c: 24 FATAL CVD: Cases: i: 1, c: 5 TOTAL MI: Cases: i: 1, c: 0NON-FATAL MI: Cases: i: 1, c: 0 TOTAL STROKE: Cases: i: 0, c: 4 NON-FATAL STROKE:	i: 36, c: 37; TOTAL CHD: Cases: i: 5, c: 11 ANGINA PECTORIS: Cases: i: 5, c: 11

	Cases: i: 0, c: 4 SUDDEN DEATH: Cases: i: 0, c: 2 REVASCULARISATION: Cases: i: 9, c: 15 HEART FAILURE: Cases: i: 4, c: 3	
Diet of intervention (i) and control (c) group	i: 1.8 g/d EPA supplementation (highly purified EPA ethyl esters) started within 24h after PCI c: nil (no placebo)	i: 2.2 g/d EPA (via 12 g/d Maxepa capsules) c: olive oil (12 g/d)
Strength of the effect: HR or RR (95%CI)	ALL-CAUSE MORTALITY: 0.22 (0.05, 1.01) TOTAL CVD: 0.46 (0.24, 0.89) FATAL CVD: 0.20 (0.02, 1.69) TOTAL MI: 3.0 (0.12, 72.91) NON-FATAL MI: 3.0 (0.12, 2.91) TOTAL STROKE: 0.11 (0.01, 2.04) NON-FATAL STROKE: 0.01 (0.00, 45.2) SUDDEN DEATH: 0.20 (0.01, 4.12) REVASCULARISATION: 0.60 (0.27, 1.32) HEART FAILURE: 0.72 (0.16, 3.24)	TOTAL CHD: 0.47 (0.18, 1.21) ANGINA PECTORIS: 0.47 (0.18, 1.21)
Study population	People having PCI after acute MI: BMI: NR; medication: aspirin (100%), statins (100%), ticlopidine (~96%), β-blockers (~60%), ARB/ACE inhibitors (~50%); men (i~77%) and women; Japan	People undergoing PTCA; medication: NR; BMI: NR; men (77%) and women; New Zealand
Compliance	NR	NR

Abbreviations: ACE: angiotensin converting enzyme; ARB: angiotensin receptor blocker; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; MI: myocardial infarction; N/A: not applicable; NR: not reported; PCI: percutaneous coronary intervention; PTCA: percutaneous transluminal coronary angioplasty; RCT: randomised controlled trial; RR: relative risk.

Footnotes:

[a] Refers to the duration of the intervention.

Table C9 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Rauch et al. (OMEGA) and Sacks et al. (HARP)

Aspect	Rauch et al. 2010[32]	Sacks et al. 1995[38]
Trial name	OMEGA	HARP
Study duration	12 months[a]	28 months (mean)[a]
Primary disease	CHD	CHD
Study design	Parallel RCT	RCT
Number of participants; health outcome; number of cases	Participants: i: 1919, c: 1885; ALL-CAUSE MORTALITY: Cases: i: 88, c: 07 TOTAL CVD: Cases: i: 534, c: 541 FATAL CVD: Cases: i: 67, c: 51 TOTAL CHD: Cases: i: 112, c: 96 FATAL CHD: Cases: i: 28, c: 29 TOTAL MI: Cases: i: 87, c: 78 FATAL MI: Cases: i: 13, c: 11 NON-FATAL MI: Cases: i: 74, c: 67 TOTAL STROKE: Cases: i: 27, c: 13 SUDDEN DEATH: Cases: i: 28, c: 29 REVASCULARISATION: Cases: i: 466, c: 482 ANGINA PECTORIS: Cases: i: 21, c: 25 HEART FAILURE: Cases: i: 467, c: 492 ARRHYTHMIA: Cases: i: 99, c: 84 CANCER: Cases: i: 32, c: 26	Participants: i: 31, c: 28; ALL-CAUSE MORTALITY: Cases: i: 0, c; 1 TOTAL CVD: Cases: i: 7, c: 7 FATAL CVD: Cases: i: 0, c: 1 FATAL CHD: Cases: i: 0, c: 1 TOTAL MI: Cases: i: 1, c: 3 FATAL MI: Cases: i: 0, c: 1 NON-FATAL MI: Cases: i: 1, c: 2 TOTAL STROKE: Cases: i: 1, c: 0 REVASCULARISATION: Cases: i: 3, c: 3 ANGINA PECTORIS: Cases: i: 3, c: 4 HEART FAILURE: Cases: i: 0, c: 1
Diet of intervention (i) and control (c) group	i: 1 g/d omega-3 acid ethyl esters (Zodin®; 460 mg/d EPA and 386 mg/d DHA) c: 1 g/d olive oil supplement	i: Fish oil capsules providing 2.88 g/d of EPA, 1.92 g/d of DHA, and 1.2 g/d of other n-3 FA (mainly DPA) c: olive oil
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	ALL-CAUSE MORTALITY: 1.23 (0.91, 1.68)	ALL-CAUSE MORTALITY: 0.30 (0.01, 7.13)

	TOTAL CVD: 0.96 (0.87, 1.06) FATAL CVD: 1.29 (0.90, 1.85) TOTAL CHD: 1.15 (0.88, 1.49) FATAL CHD: 0.95 (0.57, 1.59) TOTAL MI: 1.10 (0.81, 1.48) FATAL MI: 1.16 (0.52, 2.58) NON-FATAL MI: 1.08 (0.78, 1.50) TOTAL STROKE: 2.04 (1.06, 3.94) SUDDEN DEATH: 0.95 (0.57, 1.59) REVASCULARISATION: 0.93 (0.81, 1.08) ANGINA PECTORIS: 0.83 (0.47, 1.49) HEART FAILURE: 0.93 (0.84, 1.04) ARRHYTHMIA: 1.16 (0.87, 1.54) CANCER: 1.21 (0.72, 2.04)	TOTAL CVD: 0.95 (0.37, 2.46) FATAL CVD: 0.32 (0.01, 7.57) FATAL CHD: 0.30 (0.01, 7.13) TOTAL MI: 0.32 (0.03, 2.92) FATAL MI: 0.40 (0.01, 12.50) NON-FATAL MI: 0.48 (0.04, 5.04) TOTAL STROKE: 2.72 (0.12, 64.14) REVASCULARISATION: 0.89 (0.16, 4.83) ANGINA PECTORIS: 0.64 (0.13, 3.16) HEART FAILURE: 0.32 (0.01, 7.57) SYSTOLIC BLOOD PRESSURE: MD in change from baseline: -1.0 (-14, 12) mmHg LDL CHOLESTEROL: MD in change from baseline: 0.25 (-0.11, 0.61) mmol/L BODY WEIGHT: 2.00 (-5.43, 9.43) kg
Study population	People who had an acute MI; BMI: NR; medication: statins (94%), ACE inhibitors, β-blockers, clopidogrel and aspirin (>50%), diuretics (20-49%); men (~74%) and women; Europe	People with CHD; BMI: NR; medication in c group: β-blockers, and antiplatelet agents (>50%), calcium channel blockers, and nitrates (20-49%), ACE inhibitors, oral hypoglycaemic drugs (<20%); men (93%) and women; USA
Compliance	i: 93.1% and c: 93.2% took >70% of capsules	i: 80% and c: 90% adherence, significant levels of adipose n-3 FAs in i group (capsule count and serum level measurements)

Abbreviations: ACE: angiotensin converting enzyme; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; DPA: docosapentaenoic acid; EPA: eicosapentaenoic acid; FA: fatty acids; g/d: grammes per day; HARP: Harvard Atherosclerosis Reversibility Project; HR: hazard ratio; i: intervention group; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; NR: not reported; RCT: randomised controlled trial; RR: relative risk; USA: United States of America.

Footnotes:

[a] Refers to the duration of the intervention.

Table C10 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Singh et al. (IEIS-4) and Souied et al. (NAT2)

Aspect	Singh et al. 1997[26]	Souied et al. 2013[42]
Trial name	IEIS-4	NAT2
Study duration	1 year[a]	36 months[b]
Primary disease	CHD	Total CVD
Study design	Parallel RCT	Parallel RCT
Number of participants; health outcome; number of cases	Participants: i: 122, c: 118; ALL-CAUSE MORTALITY: Cases: i: 14, c: 26 TOTAL CVD: Cases: i: 30, c: 41 NON-FATAL CVD: Cases: i: 16, c: 30 TOTAL CHD: Cases: i: 30, c: 41 FATAL CHD: Cases: i: 14, c: 26 FATAL MI: Cases: i: 12, c: 18 NON-FATAL MI: Cases: i: 16, c: 30 SUDDEN DEATH: Cases: i: 2, c: 8 ANGINA PECTORIS: Cases: i: 22, c: 50	Participants: i: 150, c: 150; ALL-CAUSE MORTALITY: Cases: i: 3, c: 6 TOTAL STROKE: Cases: i: 0, c: 1 LDL CHOLESTEROL: Participants: i: 134, c: 129
Diet of intervention (i) and control (c) group	i: 6 g/d EPA/DHA fish oil supplements (Maxepa) containing 1.08 g/d EPA and 0.72 DHA c: placebo supplements containing 100 mg/d aluminium hydroxide	i: 1110 mg/d n-3 FAs (270 mg EPA and 840 mg DHA); c: olive oil (1800 mg/d)
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	ALL-CAUSE MORTALITY: 0.50 (0.30, 0.90) TOTAL CVD: 0.71 (0.48, 1.05) NON-FATAL CVD: 0.44 (0.23, 0.86) TOTAL CHD: 0.70 (0.29, 0.90) FATAL CHD: 0.52 (0.29, 0.95) FATAL MI: 0.60 (0.30, 1.30) NON-FATAL MI: 0.52 (0.30, 0.90)	ALL-CAUSE MORTALITY: 0.50 (0.13, 1.96) TOTAL STROKE: 0.33 (0.01, 8.12) LDL CHOLESTEROL: -0.06 (-0.30, 0.18) mmol/L

	SUDDEN DEATH: 0.23 (0.05, 1.10) ANGINA PECTORIS: 0.30 (0.17, 0.54)	
Study population	People with AMI; BMI: NR; medication: β-blockers (29%), calcium channel blockers (21%), nitrates (67%), aspirin (75%), streptokinase (5%); men; India	People with early AMD (85% had CVD); BMI: NR; medication: lipid-lowering agents (>50%); men (35%) and women; Europe
Compliance	NR (assessed by capsule count)	Overall compliance i: 69.4%, c: 70.5% (assessed from unused capsules)

Abbreviations: AMD: age-related macular degeneration; AMI: acute myocardial infarction; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; EPA: eicosapentaenoic acid; FA: fatty acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; IEIS-4: Indian Experiment of Infarct Survival-4; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; NAT2: Nutritional AMD treatment-2; NR: not reported; RCT: randomised controlled trial; RR: relative risk.

Footnotes:

[a] Mean or median follow-up not reported.

[b] Refers to the duration of the intervention.

Table C11 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCTs by Svensson et al. (OPACH) and Von Schacky et al. (SCIMO)

Aspect	Svensson et al. 2006[46]	Von Schacky et al. 1999[48]
Trial name	OPACH	SCIMO
Study duration	2 years (maximum)	24 months[a]
Primary disease	Total CVD	CHD
Study design	Parallel RCT	Parallel RCT
Number of participants; health outcome; number of cases	Participants: i: 103, c: 103; ALL-CAUSE MORTALITY: Cases: i: 34, c: 30 TOTAL CVD: Cases: i: 62, c: 59 TOTAL CHD: Cases: i: 7, c: 16 TOTAL MI: Cases: i: 4, c:13 TOTAL STROKE: Cases: i: 12, c: 5 REVASCULARISATION: Cases: i: 3, c: 4	Participants: i: 112, c: 111; ALL-CAUSE MORTALITY: Cases: i: 1, c: 2 TOTAL CVD: Cases: i: 17, c: 26 FATAL CVD: Cases: i: 0, c: 1 NON-FATAL CVD: Cases: i: 2, c: 6 TOTAL CHD: Cases: i: 1, c: 4 FATAL CHD: Cases: i: 0, c: 1 TOTAL MI: Cases: i: 1, c: 4

		FATAL MI: Cases: i: 0, c: 1 NON-FATAL MI: Cases: i: 1, c: 3 TOTAL STROKE: Cases: i: 1, c: 3 REVASCULARISATION: Cases: i: 22, c: 24 ANGINA PECTORIS: Cases: i: 9, c: 11 SYSTOLIC BLOOD PRESSURE: Participants: i: 112, c: 111 LDL CHOLESTEROL: Participants: i: 87, c: 84 BODY WEIGHT: Participants: i: 89, c: 86
Diet of intervention (i) and control (c) group	i: 1.7 g/d of n-3 PUFA (Omacor®; 0.77 g EPA + 0.64 g DHA) c: olive oil	i: fish oil capsules providing 4 g/d EPA+DHA+DPA+ALA in first 3 months, 2 g/d rest of trial (mean: 2.3 g/d) c: capsules containing fat replication of fat composition of average European diet
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	ALL-CAUSE MORTALITY: 1.13 (0.75, 1.70) TOTAL CVD: 1.05 (0.84, 1.32) TOTAL CHD: 0.40 (0.17, 0.97) TOTAL MI: 0.31 (0.10, 0.91) TOTAL STROKE: 2.40 (0.88, 6.57) REVASCULARISATION: 0.73 (0.16, 3.25)	ALL-CAUSE MORTALITY: 0.50 (0.05, 5.39) TOTAL CVD: 0.65 (0.37, 1.13) FATAL CVD: 0.33 (0.01, 8.02) NON-FATAL CVD: 0.32 (0.06, 1.61) TOTAL CHD: 0.25 (0.03, 2.18) FATAL CHD: 0.33 (0.01, 8.02) TOTAL MI: 0.25 (0.03, 2.18) FATAL MI: 0.50 (0.01, 14.70) NON-FATAL MI: 0.33 (0.03, 3.13) TOTAL STROKE: 0.33 (0.03, 3.13) REVASCULARISATION: 0.89 (0.46, 1.70) ANGINA PECTORIS: 0.81 (0.35, 1.88)

		SYSTOLIC BLOOD PRESSURE: MD in change from baseline: -0.10 (-4.90, 4.70) mmHg LDL CHOLESTEROL: MD in change from baseline: 0.16 (-0.17, 0.49) mmol/L BODY WEIGHT: -1.90 (-5.57, 1.77) kg
Study population	People with established CVD and treated with stable haemodialysis for at least 6 months; medication: statins (20%), men (65%) and women, Europe	People with CHD; BMI: NR; medication: statins (26%), platelet inhibitors and β-blockers (>50%), ACE inhibitors, diuretics, calcium antagonists and other antihypertensive agents (20-49%), nitrates only on demand (<20%); men (~80%) and women; Europe
Compliance	Compliance was evaluated by measuring serum phospholipid fatty acid composition of n-3 PUFA. Compliance results are unclear.	Erythrocyte phospholipids rose from 4.6% to 11.8% at 24 months in intervention, and did not alter from baseline in controls.

Abbreviations: ACE: angiotensin converting enzyme; ALA: alpha linoleic acid; ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; DHA: docosahexaenoic acid; DPA: docosapentaenoic acid; EPA: eicosapentaenoic acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; LA: linoleic acid; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; N/A: not applicable; NR: not reported; PUFA: polyunsaturated fatty acid; RCT: randomised controlled trial; RR: relative risk; SCIMO: Study on Prevention of Coronary Atherosclerosis by Intervention with Marine Omega-3 Fatty Acids.

Footnotes:

[a] Refers to the duration of the intervention.

Table C12 Summary of study characteristics and effects of EPA and DHA supplementation on the risk of health outcomes in people with ASCVD: individual RCT by Yokoyama et al. and Tanaka et al. (JELIS)

Aspect	Yokoyama et al. 2007[23] and Tanaka et al. 2008[40]
Trial name	JELIS
Study duration	5 years (median: 4.6 years)
Primary disease	CHD
Study design	Parallel RCT (open-label)
Number of participants; health outcome; number of cases	Participants: i: 1823, c: 1841; TOTAL CVD: Cases: i: 158, c: 197 TOTAL CHD: Cases: i: 158, c: 197

	FATAL CHD: Cases: i: 21, c: 18 NON-FATAL CHD: Cases: i: 145, c: 178 TOTAL MI: Cases: i: 42, c: 31 FATAL MI: Cases: i: 8, c: 5 NON-FATAL MI: Cases: i: 38, c: 26 TOTAL STROKE: Cases: i: 33, c: 48 SUDDEN DEATH: Cases: i: 13, c: 13 REVASCULARISATION: Cases: i: 127, c: 148 ANGINA PECTORIS: Cases: i: 88, c: 123 SYSTOLIC BLOOD PRESSURE and LDL CHOLESTEROL: Participants: i: 485, c: 457
Diet of intervention (i) and control (c) group	i: 1.8 g/d EPA supplementation (highly purified ethyl esters) c: no supplement
Strength of the effect: HR or RR (95%CI), or MD (95%CI)	TOTAL CVD: 0.79 (0.64, 0.99) TOTAL CHD: 0.81 (0.66, 1.00) FATAL CHD: 0.87 (0.46, 1.62) NON-FATAL CHD: 0.82 (0.66, 1.02) TOTAL MI: 0.75 (0.47, 1.19) FATAL MI: 0.64 (0.21, 1.94) NON-FATAL MI: 0.70 (0.42, 1.14) TOTAL STROKE: 0.80 (0.64, 1.00) SUDDEN DEATH: 1.01 (0.47, 2.18) REVASCULARISATION: 0.86 (0.67, 1.10) ANGINA PECTORIS: 0.71 (0.53, 0.94) SYSTOLIC BLOOD PRESSURE:

	MD in change from baseline: 0.0 mmHg (-2.60, 2.60) LDL CHOLESTEROL: MD in change from baseline: 0.70 mg/dL (-2.50, 4.10)
Study population	People with CHD[a]; BMI[b]: 24± 3 kg/m^2; medication: statins (100%), antiplatelet agent (42%), calcium antagonist (50%), β-blocker (18%), other hypertensive agents (35%), nitrate (38%), hypoglycaemic agents (16%); men and women; Asia (Japan)
Compliance	Unclear (similar proportions of participants remained compliant in each treatment group)

Abbreviations: ASCVD: atherosclerotic cardiovascular disease; BMI: body mass index; c: control group; CHD: coronary heart disease; CI: confidence interval; CVD: cardiovascular disease; EPA: eicosapentaenoic acid; g/d: grammes per day; HR: hazard ratio; i: intervention group; JELIS: Japan EPA Lipid Intervention Study; LDL: low-density lipoprotein; MD: mean difference; MI: myocardial infarction; N/A: not applicable; NR: not reported; RCT: randomised controlled trial; RR: relative risk.

Footnotes:

[a] Approximately 80% of the population had angina pectoris, 30% had a MI and 25% underwent coronary artery bypass graft surgery or percutaneous transluminal coronary angioplasty.

[b] Mean (± standard deviation).

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