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Review Article

Update on the omega-3 fatty acid trial landscape: A narrative review with implications for primary prevention

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Abstract: Residual risk mediated by hypertriglyceridemia among statin-treated individuals is an important clinical and public health challenge. Niacin, fibrates and omega-3 FA are three classes of non-statin agents with demonstrated TG-lowering effects. Randomized controlled trials of niacin and fibrates have been consistently negative, but the trial landscape for two key sources of omega-3 FAs, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) is more complex. Clinical trials evaluating omega-3 FA can be differentiated into those that studied mixed formulations (EPA + DHA) and those that studied EPA alone. Those assessing the impact of mixed formulations have not consistently demonstrated CVD risk reduction, whereas trials of EPA alone have been successful. Two recent trials of mixed formulations - STRENGTH (Long-Term Outcomes Study to Assess Statin Residual Risk with Epanova in High Cardiovascular Risk Patients with Hypertriglyceridemia) and OMEMI (Omega-3 fatty acids in Elderly patients with Myocardial Infarction) – studied contemporarily treated patients with mixed EPA + DHA formulations at higher doses than before and showed no benefit, thus adding valuable information to our overall understanding of this evolving therapeutic class. In this review, we contextualize the findings of STRENGTH and OMEMI within the existing omega-3 FA clinical trial landscape and look ahead to how future trials can inform existing knowledge gaps, particularly with regards to the applicability of these agents within the primary prevention realm.

Introduction

Hypertriglyceridemia and residual risk

Hypertriglyceridemia results from increases in triglyceride-rich lipoproteins such as chylomicrons and VLDL and is associated with increased ASCVD risk independent of LDL-C control.[1–3] The epidemiologic data supporting this association are robust and continue to grow, with novel observations demonstrating that apo-B containing TG-rich remnants (or VLDL) account for the majority of MI risk associated with apoB-containing lipoproteins.[4]

Abbreviations: LDL-C, Low density lipoprotein cholesterol; VLDL-C, Very low density lipoprotein cholesterol; ASCVD, atherosclerotic cardiovascular disease; CVD, cardiovascular disease; CV, cardiovascular; MI, myocardial infarction; TG, triglycerides; FA, fatty acids; Apo-B, apolipoprotein-B; AF, atrial fibrillation; HF, heart failure; LVEF, left ventricular ejection fraction; DM, diabetes mellitus; hsCRP, high sensitivity C-reactive protein; IVUS, intravascular ultrasound; CI, confidence interval; HR, hazard ratio; ITT, intention to treat.

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Further, several Mendelian randomization studies have now firmly established the genetic link between overall TG levels and ASCVD risk.[5–7] Residual risk mediated by hypertriglyceridemia among statin-treated individuals remains a vexing clinical problem with significant economic consequences.[8]

Three classes of non-statin agents with demonstrated TG-lowering effects include niacin, fibrates, and omega-3 FA. To date, large outcomes trials evaluating both niacin and fibrates as adjunctive therapy to statins have failed to demonstrate significant ASCVD risk reduction.[9,10] Two key sources of omega-3 FA, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), have been hypothesized to confer ASCVD risk reduction benefit through pleiotropic mechanisms beyond TG-lowering.[11,12]

Clinical trials evaluating omega-3 FA can be differentiated into those that studied mixed formulations (EPA + DHA) and those that studied EPA alone. No trials of DHA monotherapy have been performed. Trials assessing the impact of mixed formulations have not consistently demonstrated CV risk reduction, whereas trials of EPA alone have been successful (Table 1). In 2020, the results of two outcomes trials of mixed formulation EPA+DHA, STRENGTH (Long-Term Outcomes Study to Assess Statin Residual Risk with Epanova in High Cardiovascular Risk Patients with Hypertriglyceridemia) and OMEMI (Omega-3 fatty acids in Elderly patients with Myocardial Infarction), were simultaneously presented and published at the AHA scientific sessions.[13,14]

The purpose of this review is to contextualize the results of these trials within the broader landscape of omega-3 FA-based therapies, namely the landmark REDUCE-IT (Reduction of Cardiovascular Events with Icosapent Ethyl–Intervention Trial) trial.[15] We draw specific attention to the formulation, dosage, and patient population that appears to derive the most benefit from this evolving class of therapeutics. We also look ahead and discuss future directions for the potential applicability of these agents within the primary prevention realm (Fig. 1).

Understanding the clinical trial landscape

Trial of EPA alone: JELIS and REDUCE-IT

Recent outcomes trials using combination EPA/DHA compounds (ORIGIN [Outcome Reduction with an Initial Glargine Intervention], ASCEND [A Study of Cardiovascular Events in Diabetes], VITAL [Vitamin D and Omega-3 Trial] and the two studies that are focus of this review, STRENGTH and OMEMI) have failed to demonstrate a CV benefit.[13,14,16–18] In contrast, first JELIS (Japan EPA Lipid Intervention Study), a randomized open-label trial, and more recently REDUCE-IT, a randomized placebo-controlled trial, both tested an EPA ethyl ester compound (icosapent ethyl; IPE) at doses of 1.8 g per day in JELIS and 4 g per day in REDUCE-IT and demonstrated net benefit.[15,19]

JELIS did not specifically enroll participants with elevated TG and was anachronistic in terms of contemporary lipid-lowering guidelines (median attained LDL-C of 136 mg/dL using predominantly low-intensity statin therapy), but these issues notwithstanding, a substantial (19%) relative risk reduction was observed in major coronary events. Thus, JELIS provided the first insight that perhaps the benefit of omega-3 FA based therapies are EPA-specific, as well as the scientific rationale upon which the REDUCE-IT trial was conducted nearly a decade later. REDUCE-IT enrolled 8,179 participants with either established ASCVD or DM with other risk factors, and over 5 years follow-up, reported significant absolute risk reductions in both the overall primary (4.8%) and key secondary (3.6%) endpoints, with corresponding numbers needed to treat of 21 and 28, respectively.[15] These were striking findings that were homogenous across a broad range of baseline TG levels and were the premise upon which the FDA expanded the indication for use of IPE to include patients with elevated TG (≥150 mg/dL) and established ASCVD or DM and two or more additional ASCVD risk factors.[15,20] The rapid development of a novel class of therapeutics, however, can outpace a comprehensive understanding of the underlying biology. This may be the case with both EPA and DHA, although both STRENGTH and OMEMI provide several relevant clinical insights which will now be discussed in turn.

The STRENGTH trial

STRENGTH was a double-blind, randomized, multicenter trial involving 13,078 statin-treated participants (median LDL-C 75 mg/dL) with high CV risk (56% with established ASCVD, 70% with DM), TG ≥180 to 500 mg/dL (median 240 mg/dL) and low HDL-C (median 36 mg/dL), designed to study a carboxylic acid formulation of EPA and DHA (omega-3 CA) against a corn oil placebo. The carboxylic acid formulation has garnered traction given its enhanced oral bioavailability compared to the EPA ethyl ester formulation utilized in REDUCE-IT.

Enrollment in STRENGTH occurred between 2014 and 2017, and the primary measure of efficacy was a composite endpoint of CV death, non-fatal MI, non-fatal stroke, and coronary revascularization or unstable angina requiring hospitalization. The Data Safety and Monitoring Committee halted the study in early January 2020 when 1,384 primary endpoints had been recorded (after median follow-up of 39 months) due to a low probability of demonstrating a net clinical benefit of omega-3 CA compared with corn oil placebo.

The primary composite endpoint occurred in 12.0% of patients treated with omega-3 CA and 12.2% of patients treated with corn oil (hazard ratio [HR] 0.99, 95% CI 0.90-1.09, P=0.84). The secondary endpoint of CV death, MI or stroke occurred in 8.3% of patients treated with omega-3 CA and 7.9% of patients treated with corn oil (HR 0.91, 95% CI 0.81-1.02, P=0.09). All-cause mortality occurred in 5.7% of patients in the omega-3 CA group and 5.1% in the corn oil group (HR 1.13, 95% CI 0.97-1.31, P=0.11). Furthermore, when stratified into primary and secondary prevention groups, the event rate was numerically reduced in only the secondary prevention arm, with the HR for the primary composite outcome of 0.94 (95% CI 0.84-1.05) in the secondary

Table 1 Summary of contemporary omega-3 FA trials categorized into those that evaluated combination formulations (EPA+DHA) and those that studied purified EPA alone. Combination EPA+DHA trials are further categorized into those that studied a high risk, post-MI, secondary prevention population versus patients with DM but no established ASCVD versus patients with established heart failure. Omega-3 FA formulation as either ethyl esters or carboxylic acids denoted when specified. Note wide variability in baseline statin use among these studies: GISSI-P: <5%; OMEGA: 94%; Alpha Omega: 86%; ORIGIN: 54%; ASCEND: 75%; VITAL: 35%. GISSI Heart Failure is Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto miocardico-Heart Failure. Other trial expansions as per primary text.

Cardiovascular outcomes trials of mixed omega-3 fatty acids

Trial	Treatment	Comparison	Patient Population	Primary Endpoint	Primary Finding
Trials of EPA+DHA post-MI					
GISSI Prevenzione[25]	850-882 mg EPA+DHA as ethyl esters in average EPA/DHA ratio of 1:2, and 300 mg vitamin E (alone and in combination)	No supplementation	11323 patients w/ recent (≤3 months) MI	Composite of all-cause mortality, nonfatal MI, stroke	EPA+DHA significantly reduced primary endpoint by 15% (95% CI 2-25%, P=0.023) over 3.5 years follow-up
OMEGA[21]	1 g gelatin capsule w/ 460 mg EPA + 380 mg DHA as ethyl esters	1 g olive oil	3851 patients ≤14 days of acute MI	Sudden cardiac death; composite secondary endpoint of all-cause mortality and nonfatal CV events	No significant impact of EPA+DHA supplementation on primary or secondary endpoints added to usual care at 1-year follow-up
Alpha Omega[23]	~400 mg of EPA + DHA in margarine (also studied margarine with EPA+DHA+ALA)	Placebo (margarine w/ no omega-3 FA added)	4837 patients w/ history of MI on contemporary secondary prevention therapies	Composite MACE endpoint comprised fatal and nonfatal CV events and cardiac interventions	Neither EPA+DHA alone nor in combination w/ ALA reduced primary endpoint (HR 1.01, 95% CI 0.87-1.17, P=0.93)
SU.FOL.OM3[22]	Daily dietary supplement containing 5-methyltetrahydrofolate (560 μg), vitamin B-6 (3 mg), and vitamin B-12 (20 μg) or placebo; and containing omega-3 FA (600 mg of EPA + DHA acid at a ratio of 2:1) or placebo	Placebo (formulation not specified)	2501 patients w/ history of MI, unstable angina, or ischemic stroke	Major CV events, defined as composite of non-fatal MI, stroke, or death from CV cause	Allocation to omega-3 FA had no significant effect on major vascular events (HR 1.08, 95% CI 0.79-1.47, P=0.64)
OMEMI[14]	1.8 g omega-3 FA capsules of 930 mg EPA + 660 mg DHA	Corn oil placebo	1027 elderly patients w/ recent AMI (≤8 weeks)	Primary composite endpoint of non-fatal AMI, unscheduled revascularization, stroke, death, HF hospitalization; secondary endpoint of incident AF	No significant reduction in primary endpoint at 2 years follow-up; numeric increase in new AF in omega-3 FA group (7.2% vs 4.0%, HR 1.84, 95% CI 0.98-3.45, P=0.06)
Trials of EPA+DHA in DM and high risk primary prevention					
ORIGIN[18]	1 g of omega-3 FA containing 465 mg EPA + 375 mg DHA as ethyl esters	1 g olive oil	12536 patients at high ASCVD risk w/ DM or impaired fasting glucose	Primary outcome of death from CV causes	No reduction in primary outcome w/ omega-3 FA (HR 0.98, CI 0.87-1.10, P=0.72); no reduction in individual components of endpoint (major vascular events, all-cause death, arrhythmic death)

(*continued on next page*)

Table 1 (*continued*)

Cardiovascular outcomes trials of mixed omega-3 fatty acids

Trial	Treatment	Comparison	Patient Population	Primary Endpoint	Primary Finding
ASCEND[16]	1 g capsules containing 840 mg of omega-3 FA (460 mg EPA + 380 mg DHA)	1 g olive oil	15480 patients w/ DM (median 7 years duration) but w/out established ASCVD	Composite of first serious vascular event (nonfatal MI/stroke, TIA, vascular death); secondary endpoint of first serious vascular event or any major revascularization	No significant difference in risk of serious vascular events w/ omega-3 FA supplementation vs placebo at median 8 year follow-up
VITAL[17]	Vitamin D plus omega-3 FA (1 g capsules containing 465 mg EPA + 375 mg DHA)	Placebo (formulation not specified)	24871 patients w/out established ASCVD	Primary composite endpoints of MACE (MI, stroke, CV mortality) and invasive cancer	Supplementation omega-3 FA did not reduce MACE events or incident cancer
STRENGTH[13]	4g omega-3 carboxylic acid formulation of 550 mg EPA + 200 mg DHA formulated as free fatty acid	Corn oil placebo	13078 statin-treated patients w/ atherogenic dyslipidemia and high ASCVD risk (44% primary prevention, 56% secondary prevention)	Primary outcome composite of CV death, non-fatal MI, non-fatal stroke, coronary revascularization or unstable angina	Trial halted prematurely by DSMB when 1384 patients experienced primary endpoint due to low probability of demonstrating clinical benefit
Trials of EPA+DHA in heart failure					
GISSI Heart Failure[48]	850-882 mg EPA+DHA as ethyl esters in average EPA/DHA ratio of 1:2, and 10 mg rosuvastatin added to usual care	Placebo (formulation not specified)	7046 patients w/ chronic HF (NYHA class II-IV); mean LVEF 33%	ITT analysis of time to death; combined secondary endpoint of time to death or HF hospitalization	Significantly reduced all-cause mortality in EPA+DHA group (HR 0.91, 95% CI 0.83-0.99, P=0.04) and combined endpoint (HR 0.92, 95% CI 0.85-0.99, P=0.01) NNT of 44 to avoid one death of HF admission over 4 years follow-up
Trials of EPA alone					
JELIS[19]	1800 mg per day of highly purified (>98%) EPA ethyl ester (w/ statin)	Statin alone	18645 Japanese patients w/ hypercholesterolemia (80% primary prevention, 20% secondary prevention)	Any major coronary event, including sudden cardiac death, fatal and nonfatal MI, and other nonfatal events including unstable angina and unplanned revascularization	19% relative risk reduction in primary composite endpoint in EPA-treated group vs control group (p=0•048). Numeric but non-significant reduction in events in primary prevention group
REDUCE-IT[15]	4g daily icosapent ethyl (purified EPA ethyl ester); 960 mg EPA per g of IPE	Mineral oil placebo	8179 patients w/ established ASCVD or with DM and other risk factors on statin-therapy w/ TG 135-499 mg/dL and LDL-C 41-100 mg/dL (29% primary prevention, 71% secondary prevention)	Composite of CV death, nonfatal MI, nonfatal stroke, coronary revascularization, or unstable angina; Secondary end point composite of CV death, nonfatal MI, or nonfatal stroke	Significantly reduced incidence of primary endpoint in IPE group (HR 0.75, 95% CI 0.68-0.83; P<0.001) Significantly lower rate of CV death in IPE group (HR 0.80, 95% CI 0.66-0.98, P=0.03) No significant interaction between treatment effect and primary vs secondary prevention strata (P for interaction=0.14)

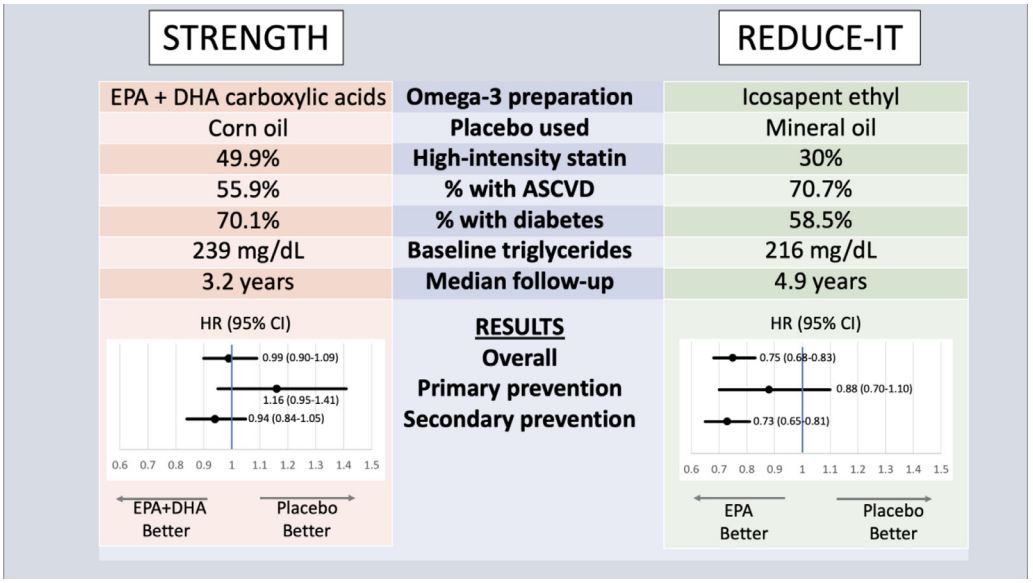

Fig. 1 Differences between the STRENGTH and REDUCE-IT trial in terms of design, patient characteristics and key results. Hazard ratios and 95% confidence intervals shown refer to the primary composite endpoint for each trial, comprising the composite of cardiovascular death, non-fatal myocardial infarction, non-fatal stroke, and coronary revascularization or unstable angina requiring hospitalization. Results of subgroup analyses by primary and secondary prevention strata are also shown.

prevention arm versus 1.16 (95% CI 0.95-1.41; P for interaction 0.06) in the primary prevention arm.

There was greater reduction in TG (-19.0 vs -0.9%, P<0.001), non-HDL-C (-6.1 vs -1.1%, P<0.001) and hsCRP (-20.0 vs -6.3%, P<0.001) in the omega-3 CA treatment group compared with corn oil. With regard to safety and tolerability, there was an excess rate of gastrointestinal adverse events in the omega-3 CA group (24.7%) versus corn oil-treated patients (14.7%) and an increased rate of new onset AF (2.2 vs 1.3%, HR 1.69, 95% CI 1.29-2.21, P<0.001). Importantly, despite the premature study termination, the number of adjudicated primary endpoint events was in keeping with the original sample size assumptions, thus obviating concern for lack of statistical power in the interpretation of the results. A comparison of results for biomarkers and endpoints in STRENGTH and REDUCE-IT is provided in Table 2.

The OMEMI trial

The OMEMI trial was a multi-center, randomized control trial conducted in Norway that studied the utility of a combination omega-3 FA formulation (930 mg EPA and 660 mg DHA) versus a corn oil placebo among elderly individuals (aged 70-82 years) with recent (within 8 weeks) MI. Importantly, the study question was fundamentally different from JELIS, REDUCE-IT and STRENGTH, as it was conceived to help inform pre-existing knowledge gaps surrounding not only the benefit of combination EPA+DHA compounds, but also the utility of omega-3 FA therapies in the post-MI period and in the elderly.[21–23] The primary measure of efficacy was the composite endpoint of non-fatal MI, unscheduled revascularization, stroke, HF hospitalization, and all-cause mortality. Unlike prior studies of omega-3 FA, OMEMI was powered to study incident AF as a secondary endpoint, and not just a safety signal.

OMEMI was a considerably smaller study (which ultimately proved underpowered) compared to STRENGTH and REDUCE-IT, randomizing 1,027 participants, in whom mean age was 75±3.6 years and 294 (29%) were female. Overall CV risk was high, with 467 (46%) having established ASCVD and 350 (35%) having prior coronary arterial stenting or bypass grafting. The vast majority (97%) were receiving statin therapy at baseline, and mean LDL-C was 75.1±25.9 mg/dL, HDL-C, 49.3±15.2 mg/dL and TG, 115.4±72.1 mg/dL. At the time of randomization, 255 (25.1%) participants had experienced some form of AF. A substantial (41%) proportion of participants reported use of non-prescription omega-3 FA supplement use at baseline.

The primary composite outcome occurred in 21.4% participants in the treatment arm and 20% in the placebo arm (HR 1.08, 95% CI 0.82-1.41, P=0.60). Additionally, no significant difference was observed in terms of any of the individual components of the composite outcome. Although not achieving statistical significance, there were numerically more new AF events in the omega-3 FA arm (7.2%) versus placebo (4.0%), (HR 1.84, 95% CI 0.98-3.45, P=0.06). Importantly, the treatment effect on the primary outcome did not differ across strata of baseline TG level or a range of other subgroups, including those with DM, previous MI, or use of omega-3 FA supplementation at baseline.

There was greater reduction in TG (-8.1 vs +5.1%, P<0.001) but no significant difference in LDL-C changes (+0.1% vs +0.7%, P=0.57) in the omega-3 FA treatment group compared with corn oil. At 2 year follow-up, median changes in serum EPA and DHA concentrations were +87% and +16% in the omega-3 FA group versus -13% and -8%

Table 2 Biochemical and cardiovascular endpoints in STRENGTH and REDUCE-IT. LDL-C, low-density lipoprotein cholesterol. EPA, eicosapentaenoic acid. *EPA concentrations reported as plasma levels in STRENGTH and serum levels in REDUCE-IT. [a]Data are presented as annualized event-rates, and for REDUCE-IT, are estimated from published Kaplan Meier Plots. The primary endpoint in REDUCE-IT and STRENGTH was similar, comprising the composite of cardiovascular death, non-fatal myocardial infarction, non-fatal stroke, and coronary revascularization or unstable angina requiring hospitalization.

Biochemical Parameters

	REDUCE-IT Icosapent Ethyl	STRENGTH Omega-3 CA	REDUCE-IT Mineral Oil	STRENGTH Corn Oil
Baseline Triglycerides, mg/dL	217	239	216	240
Month 12, median	175	191	221	235
Change, median	-39	-42	4.5	-2
Percent change, median	-18	-19	2.2	-0.9
Baseline LDL-C, mg/dL	74	75	76	75
Month 12, median	77	76	84	75
Change, median	2	1.0	7	-1.0
Percent change, median	3.1	1.2	10.2	-1.1
Baseline Plasma EPA, ug/mL*	26	21	26	21
Month 12, median	144	89.6	23	19
Percent change, median	394	269	-12.8	-10.5

Cardiovascular Event Rates[a]

	REDUCE-IT Icosapent Ethyl	STRENGTH Omega-3 CA	REDUCE-IT Mineral Oil	STRENGTH Corn Oil
Primary Endpoint				
1 years	4.5	3.8	5.0	3.9
2 years	8.5	7.0	10.0	7.2
3 years	13.0	10.3	16.0	10.3
4 years	16.0	13.1	20.0	13.6

in the corn oil group, respectively. With regard to safety and tolerability, major bleeding occurred in 10.7% in the omega-3 FA group and 11.0% in the placebo group, but importantly no participants withdrew from the trial due to bleeding problems.

In addition to small sample size, other important limitations of OMEMI include a relatively short follow-up period for a CV outcomes trial and the fact that baseline levels of phospholipid EPA and DHA were consistently higher than those observed in the contemporary North American population, likely owing to the fact that participants were allowed to consume supplemental cod liver oil for the duration of the trial. These limitations notwithstanding, the findings from OMEMI inform knowledge gaps existing the wake of early randomized control trials from the 1990s such as DART (The Diet and Reinfarction Trial) and GISSI-P (Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto miocardico-Prevenezione) which suggested a CV benefit to omega-3 FA based therapies following MI.[24,25] The enthusiasm generated by these two studies was ultimately quelled nearly a decade later with the publication of three large trials – OMEGA-Study (Effect of Omega 3-Fatty Acids on the Reduction of Sudden Cardiac Death After Myocardial Infarction), Alpha Omega and SU.FOL.OM3 (Supplementation with Folate, vitamin B6 and B12 and/or Omega-3 fatty acids) – that studied combination EPA and DHA formulations doses ranging from 400 – 840 mg/day, all of which reported no benefit (Table 1).[21,22]

By contrast, OMEMI studied an older, higher risk population treated with contemporary post-MI therapies (including dual-antiplatelet therapy and high-intensity statin) and employed a higher dose of omega-3 FA than previously studied, thus filling an important knowledge gap. Taken together, these four studies provide strong evidence that in a very high risk secondary prevention setting, combination EPA/DHA therapy renders no significant CV benefit across a spectrum of dosages, a message that appeared consistent across analyses of various subgroups and individual components of the primary outcome in OMEMI.

Following the publication of STRENGTH and OMEMI, members of the scientific community expressed concern that the findings of these trials provide some uncertainty regarding the clinical utility of omega-3 FA for the prevention of CV disease. There are a number of issues raised warranting further discourse, in particular as the results of STRENGTH are juxtaposed against those from REDUCE-IT, and these will be addressed in turn.

Comparing strength & OMEMI to reduce-it

Drug composition

The foremost difference to address relates to the composition of study drug used in each trial. Whereas JELIS and REDUCE-IT administered an EPA ethyl ester compound,

STRENGTH used a combination of EPA and DHA. The formulation studied in REDUCE-IT contained 960 mg EPA per capsule, while the formulation in STRENGTH contained 550 mg EPA and 200 mg DHA per capsule. As mentioned, combination therapy was also employed in VITAL, ASCEND, and the GISSI trials preceding them.[16,18,25]

A key structural difference between EPA and DHA is that DHA has an additional double bond and two more carbons compared to EPA. This impacts the nature of their discrete interaction with surrounding membrane lipids and may have implications on lipid raft formation and signal transduction pathways.[26,27] For example, compared to DHA, EPA inserts into lipoprotein particles and cellular membranes in a more extended conformation enabling it to scavenge reactive oxygen species more efficiently.[26,28] In contradistinction, DHA interacts with membrane phospholipids in a more disordered and less stable fashion, a phenomenon that likely explains its greater susceptibility towards isomerization and reduced antioxidant activity relative to EPA.[27]

These differential effects in membrane fluidity and lipid domain alterations may underpin, if at least to a minor degree, some of the discrepant outcomes observed in the clinical trial data. These mechanistic differences also may explain other important differences in the independent effects of EPA and DHA on markers of CV risk. For example, the fairly consistent observation of a dose-dependent increase specific to DHA on LDL-C and HDL-C juxtaposed against the (albeit modest) net percent decreases in LDL-C and non-HDL-C specific to EPA.[29,30] Moreover, these mechanistic differences are likely at play in considering the pleiotropic impact seemingly specific to EPA beyond lipoprotein metabolism, ranging from beneficial effects on endothelial function and oxidative stress to the mitigation of nascent and stabilization of established plaque.[28,31]

Beyond the specific differences between EPA and DHA, the formulation of drugs used in the various trials, be they ethyl esters or carboxylic acid iterations, may have importance. For instance, the carboxylic acid compound utilized in STRENGTH formulated as a free fatty acid has relevant differences in its interaction with the intestinal mucosa versus the ethyl ester formulation used in REDUCE-IT.[13,15,32] Namely, it does not require hydrolysis by pancreatic lipase during intestinal absorption, which has the benefit of enhanced oral bioavailability compared to ethyl esters and thus eliminates the need for consumption with a fatty meal.

This perceived benefit notwithstanding, the rapid interaction of the free fatty acid formulation with the intestinal mucosa (as compared with the slower, more controlled release of fatty acids resulting from intestinal lipolysis of esterified formulations) is a potential driver of the significant excess in gastrointestinal adverse effects reported in the treatment arm of STRENGTH relative to placebo (24.7% vs 14.7%).[13] Any impact that the free fatty acid formulation in STRENGTH had on systemic immune activation via the gut (and thus potentially, on the primary endpoints of the trial) as a result of these more rapid kinetics with the intestinal epithelium are indeed speculative, but based on the available data, the value of an adequately esterified EPA and DHA combined formulation remains uncertain. No prior study of combination EPA and DHA compounds studied a carboxylic acid formulation.

Achieved on-treatment omega-3 FA blood levels

Subsequent analyses within REDUCE-IT have demonstrated a direct relationship between the achieved on-treatment blood level of EPA and the attendant CV benefit.[33] In STRENGTH, the administration of omega-3 CA substantially raised both plasma and red blood cell membrane concentrations of EPA (269% and 299%, respectively), but importantly, not quite to the extent observed in REDUCE-IT (394% in serum). Omega-3 CA treatment also raised plasma and red blood cell membrane concentrations of DHA by 40% and 24%, respectively, while the EPA ethyl ester formulation in REDUCE-IT decreased serum levels of DHA by 3%.

Interestingly, STRENGTH reported no association between achieved on-treatment EPA or DHA levels and CV risk. It is important to note that the median on-treatment EPA concentrations in REDUCE-IT were significantly higher (135 ug/mL) than STRENGTH (89 ug/mL). Thus, it is plausible that the potential benefits of raised EPA levels in STRENGTH were offset by the substantial concomitant increase in DHA levels, thereby explaining the lack of clinical benefit in the face of substantial improvements in biochemical parameters.

A second possible consideration could be that achieved on-treatment blood levels of EPA in STRENGTH was not high enough to render significant benefit. Refuting these hypotheses to some extent is a recently published secondary analysis of the STRENGTH trial reporting that the highest achieved tertiles of EPA (HR for primary outcome 0.98; 95% CI 0.83-1.16; P=0.81) and DHA (HR for primary outcome 1.02; 95% CI 0.86-1.20; P=0.85) were associated with neither benefit nor harm, respectively.[34] A recent meta-analysis of 40 interventional trials of omega-3 FA based therapies reported that the net CV benefit is dose-dependent, lending further credence of the relevance of the achieved on-treatment concentrations.[35]

Study population

In terms of baseline patient characteristics, 71% of REDUCE-IT participants (versus 56% of STRENGTH) had established ASCVD at baseline.[15] As a corollary of this, the absolute risk reduction observed in REDUCE-IT would expectedly be more pronounced than STRENGTH, simply as a reflection of the higher baseline risk. This is further reflected in the higher event rates observed in the placebo groups of REDUCE-IT versus STRENGTH.[13,15] Also, as mentioned earlier, STRENGTH did report a trend towards a benefit in the omega-3 CA group among secondary prevention participants, while corn oil placebo appeared favorable among the primary prevention group. Thus, it is plausible, although unproven, that an overall benefit to omega-3 CA therapy may

have been observed in the context of a larger secondary prevention arm.

Another relevant consideration is the percentage of participants with DM in each trial, namely, 70.5% in STRENGTH and 41.5% in REDUCE-IT.[13,15] REDUCE-IT reported a significant benefit irrespective of DM status at baseline, compared to STRENGTH, which reported a nonsignificant trend towards benefit among diabetics only in the treatment arm.[15] The marked heterogeneity of the spectrum of subclinical atherosclerosis among those with DM must be highlighted in interpreting these results, which begets the possibility that a more precise estimate of each trial's primary prevention arm (for example, by stratification according to burden of subclinical atherosclerotic disease) could help elucidate if these participants were at comparable ASCVD risk or not.[36–38] Acknowledging that neither STRENGTH nor REDUCE-IT were powered to probe the specific question of benefit in a strictly primary or secondary prevention cohort, these differences in the baseline characteristics of the trial participants are important to reconcile.

Comparator group

The third difference between these trials is the nature of the comparator employed. STRENGTH used corn oil and REDUCE IT used mineral oil. Although a general tenet of all placebo-controlled trials is that the placebo be inert, omega-3 FA intervention trials often inherently employ biologically active oils as placebo due to limited options in the selection of colorless and odorless capsules that can mimic the active study drug in these trials.

The STRENGTH investigators highlight the use of corn oil as a neutral comparator with the least effects on a range of biochemical parameters associated with CV risk. In contrast, the mineral oil comparator employed by REDUCE-IT had demonstrable adverse effects on LDL-C (10% increase), hsCRP (32% increase) and apo-B levels (8% increase).[15] These observations notwithstanding, given the relatively widespread use of mineral oil placebo in previous clinical trials, in 2015, following rigorous investigation of the available data, the European Commission amended European Union regulations to add pharmaceutical-grade mineral oils to the list of substances/active ingredients that do not pose a significant risk.[39] The US Food and Drug Administration has likewise signed off on the use of mineral oil as an inert placebo.[40]

A recently published, systematic review of over 80 studies wherein mineral oil was utilized as placebo reported inconsistent and generally statistically insignificant changes across a range of lipid parameters including TG, LDL-C, HDL-C, and hsCRP, among other biomarkers, concluding that mineral oil use as placebo does not meaningfully affect study conclusions at the quantities used in clinical trials.[41]

Of note, the EVAPORATE (Effect of Vascepa on Improving Coronary Atherosclerosis in People With High Triglycerides Taking Statin Therapy) trial investigators compared (via post hoc analysis of observational data from two trials) the rates of plaque progression with mineral oil compared to a cellulose-based placebo, demonstrating no significant differences in progression of total plaque and non-calcified plaque.[42] This important work was the first to substantiate, on the basis of atherosclerosis imaging, that the changes observed among mineral oil-treated participants in levels of LDL-C and inflammatory markers in REDUCE-IT were unlikely to be of pathobiological consequence.

Implications for primary prevention

The clinical trials discussed thus far were designed predominantly to study the utility of omega-3 FA based therapies within the realm of secondary or high risk primary prevention populations. JELIS only demonstrated benefit in reducing atherosclerotic coronary and stroke outcomes in the secondary prevention cohort, not in the primary prevention cohort.[19] Similarly, when the REDUCE-IT study population was stratified into primary (29% participants) and secondary prevention (71% participants) cohorts, a significant benefit was only observed in the secondary prevention arm (HR 0.72, CI 0.63-0.82 versus HR 0.81, CI 0.62-1.06 for primary prevention, P-value for interaction=0.41). However, the clear signal from REDUCE-IT that the benefit of IPE is largely independent of the degree of TG-lowering supports to notion that EPA may have applicability within the broader realm of primary prevention settings. Once again, this relates to the growing body of evidence supporting the pleiotropic effects of EPA on mitigating endothelial dysfunction, inflammation and the propagation of plaque.[10,28]

Specifically, across each of these domains, EPA has been associated with increased nitric oxide bioavailability and decreased monocyte adhesion; with increased interleukin-10 and pro-resolving mediators and decreased intercellular adhesion molecule-1; and with increased fibrous cap thickness and decreased plaque volume.[12,28,43,44] In the future, outcomes trial data will be needed to better inform the baseline risk (e.g., a study of participants with DM stratified by baseline degree of subclinical atherosclerosis) and dose of EPA that may be useful in the primary prevention setting.

Within the past 5 years, two studies have employed plaque imaging to demonstrate with increased clarity the benefits of EPA on plaque stabilization and regression. The CHERRY (Combination Therapy of Eicosapentaenoic Acid and Pitavastatin for Coronary Plaque Regression Evaluated by Integrated Backscatter Intravascular Ultrasonography) trial was an IVUS study, which clearly demonstrated that the addition of EPA to high-dose pitavastatin therapy among patients with established ASCVD who had undergone recent percutaneous coronary intervention induced significant reductions in both total atheroma and lipid volume over 8 months follow-up.[45]

The more recent EVAPORATE trial was a randomized double-blind trial utilizing serial multidector computed tomography that showed a significant regression of low attenuation plaque volume at 18 months among participants

with similar baseline characteristics to REDUCE-IT treated with IPE relative to a mineral oil placebo.[46] Importantly, IPE treatment resulted in regression in both fibrous and fibrofatty plaque volumes compared to the progression experienced in the treatment group; these findings remained significant following multivariable adjustment for age, sex, DM status, hypertension and baseline TG levels. In accordance with recent calls within the scientific community to enrich primary prevention trials with atherosclerosis imaging by way of CAC, a compelling future placebo-controlled trial to further define the role of IPE in primary prevention may study IPE in individuals with CAC $\geq$100 without DM.[47]

Conclusions & future directions

The science of omega-3 FA is indeed complex, and the magnitude of CV benefit has been a point of debate for decades. The mixed results of clinical trial data stem from myriad sources, including inherent differences in the biology of EPA and DHA, the formulation of compound studied (ethyl ester versus carboxylic acid), the respective dose of EPA and DHA within the commercially available formulations, the achieved on-treatment levels of EPA and DHA, as well as intrinsic differences in trial design including the baseline risk of the participants and the challenge of identifying a suitably inert placebo. STRENGTH and OMEMI have been highly informative in adding to our collective understanding of this science.

STRENGTH shows that among high risk, statin-treated primary and secondary prevention patients with atherogenic dyslipidemia, similar to but more contemporary than those studied in ORIGIN, ASCEND and VITAL, treatment with a mixed EPA+DHA at higher doses than previously studied with a more bioavailable carboxylic acid formulation did not confer a significant CV benefit. OMEMI shows that among elderly patients with recent MI, similar to but more contemporary than those studied in GISSI-P, OMEGA, Alpha Omega, and SU.FOL.OM3, treatment with a mixed EPA+DHA also at higher doses than previously studied similarly conferred no significant CV benefit.

Thus, the CV benefit of an omega-3 FA based therapeutic strategy may be specific to whether or not EPA is used alone or combined with DHA. The results of REDUCE-IT and JELIS clearly support this. Further, this benefit appears to be independent of TG-lowering and may relate to the pleiotropy of EPA discussed herein manifest in its anti-inflammatory properties and overall beneficial effects on vascular function. Particularly in light of evidence from imaging studies that purified EPA halts the progression and indeed induces plaque regression, and due to the lack of benefit in primary prevention, a focus on plaque burden rather than primary or secondary prevention makes sense. We suggest that to determine the extent and specificity of its therapeutic utility, future studies should focus on individuals with and without DM stratified by the amount of CAC.

Authors contribution

All authors have approved the final version of this article. K.K and A.A contributed to writing and editing of the article. N.J.S and R.S.B were senior authors involved in planning, reviewing, writing, editing and producing the article.

Declarations of Competing Interest

K.K., A.A, N.J.S, and R.S.B have no conflicts of interest to declare.

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Disclosures

None.

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