Cardiac Membrane Polyunsaturated Fatty Acids as Therapeutic Targets in Age-associated Heart Diseases

a report by

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Modification of cardiac membrane lipid composition leads to functional changes in lipid–protein interactions and related metabolic processes that are a major factor underlying the reduced capacity to recover from acute injury or to adapt during chronic disease in advanced age.[1] In addition to perturbed regulation of ion homeostasis, metabolic intermediates and antioxidant systems, important adaptive and maladaptive membrane phospholipid changes occur in ageing and age-related heart disease. We have previously reported that with advanced age there are increases in the omega (ω)-6 polyunsaturated fatty acids (PUFAs) and phosphatidylcholine, in contrast to decreases in ω-3 PUFAs and cardiolipin.[2,3]

A deficiency of long-chain ω-3 PUFAs in myocardial membranes has been proposed to underlie an increased vulnerability to cardiac arrhythmias in age-related cardiovascular disease. Phospholipids and their constituent fatty acids are integral to cell membrane function and to biologically active membrane-derived signalling products, including fatty acid diacylglycerols, inositol triphosphate and fatty acid metabolites. Many PUFAs are required in the diet as mammals cannot synthesise them *de novo*.[4,5] Of the two main families, ω-6 PUFAs are readily available in the diet as linoleic acid (LA). The main PUFAs in myocardial membranes are the ω-6 PUFAs, LA and arachidonic acid (AA), and the ω-3 PUFA docosahexaenoic acid (DHA).

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With abundant LA and high levels of ω-6 PUFA in the 'modern' diet of developed nations, α-linolenic acid (ALA) cannot be converted to adequate levels of eicosapentaenoic acid (EPA) and DHA. This is due to competition for the desaturase enzymes with the vast excess of dietary ω-6 PUFA.[4–8]

Delta-5 and delta-6 fatty acid desaturases are crucial for the conversion of both ω-6 PUFAs (LA to AA) and ω-3 PUFA (ALA to EPA and DHA).[4–8] The activity of microsomal delta-6 desaturase is lower than that of delta-5, making it the rate-limiting step involved in two stages of DHA production.[8] Thus, in the face of a dietary intake that is low in ALA and high in LA and AA (e.g. meat), there is usually a reduction in ω-3 PUFA content in membranes; this is exacerbated by any downturn in desaturase enzyme function.[5,6]

Altered Response to Ischaemia-reperfusion Injury

Notably, the consequences of these dietary effects further exaggerate low ω-3 PUFA and high ω-6 PUFA content in cardiac membranes and compound the development and impact of heart disease in advanced age. In senescent rat hearts, ischaemia-reperfusion injury results in augmented Ca^{2+} accumulation in cytoplasm and mitochondria and reduced recovery of contractile and mitochondrial function compared with younger counterparts.[9,10] In human atrial trabeculae, we have also shown the impact of advanced age in reducing recovery of post-ischaemic contractile function.[11] Dietary treatment for only a few weeks with fish oil can circumvent the effects of a high-ω-6-PUFA diet in young and old adult rats because it increases the incorporation of cardiac membrane ω-3 PUFA, the recovery of contractile work, Ca^{2+} homeostasis and O_2 utilisation efficiency, and reduces the incidence of arrhythmias after acute ischaemia and reperfusion.[9,12–16]

The consequences of a decrease in the ratio of ω-3 to ω-6 PUFAs include changes in crucial membrane lipid–protein interactions that lead to altered intracellular signalling, reduced efficiency of oxygen utilisation, failing mitochondrial energy production, augmented free radical formation, lipid and protein oxidation and modification and decreased ion homeostasis.[1,17] These changes create a vicious circle that is triggered by adverse events such as ischaemic injury and oxidative stress. Notably, superoxide free radicals form ω-6 PUFA-derived cytotoxic and chemotactic reactant lipid hydroperoxides (i.e. 4-hydroxy-2-nonenal, 4-hydroxynonenal [HNE]) that interact with the sulfhydryl and thiol groups of adjacent proteins to form protein carbonyls and other structural modifications. These limit or inactivate numerous enzyme- and ion-exchange systems, creating dysfunction in energy metabolism and ion homeostasis.[1,17] Thus, high cardiac membrane ω-6 PUFA, coupled with increased reactive oxygen species (ROS) in ageing, magnifies myocardial dysfunction after adverse events such as ischaemia or in chronic conditions such as hypertrophy, heart failure or diabetes.

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Prostacyclin, a vasodilator eicosanoid, is produced from the ω-6 PUFA AA and the ω-3 PUFA EPA, whereas the active form of the vasoconstrictor, thromboxane, is produced only from the ω-6 PUFA AA. It is widely accepted that a low ratio of EPA to AA predisposes to a balance of eicosanoids favouring vasoconstriction, platelet aggregation, inflammatory mediator signalling and cardiac arrhythmias.[16,18–23] During acute stress, augmented free radical production, peroxidation of ω-6 PUFA and production of specific prostanoids, leukotrienes, cytokines and other inflammatory mediators promote metabolic, ionic and electrical instability and depress contractile dysfunction.[18–23]

Recently, it has been shown in coronary artery bypass graft (CABG) surgery patients that epicardial adipose tissue around the proximal coronary arteries is a rich source of inflammatory cytokines such as interleukin (IL)-6, IL-1β, tumour necrosis factor (TNF)-α and surgery promotes their release.[19–25] Thus, the resultant rise in ω-6 PUFA-derived products, particularly those predisposed to increase disease and surgical interventions, may be targeted by dietary intervention to augment ω-3 PUFA incorporation into tissues, diminishing the excess of ω-6 PUFA-derived metabolites.

Despite recent controversies in clinical studies, there are considerable clinical data that support the extensive animal data that have demonstrated a specific antiarrhythmic action of dietary fish oil.

Correcting Omega-3 Polyunsaturated Fatty Acid Deficits

Rodent and primate hearts are responsive to increases in dietary ω-3 PUFA intake and preferentially accumulate DHA well above the circulating levels even after feeding pure EPA.[15,16,27] Notably, dietary supplementation with ω-3 PUFA can reverse the age-associated decline in ω-3 PUFA and accumulation of ω-6 PUFA in cardiac membranes. This has important implications in disease states that are more common in advanced age. After ischaemia-reperfusion, ω-3 PUFA treatment is associated with markedly reduced vulnerability to arrhythmia, improved pumping and O_2 utilisation efficiency and maintenance of Ca^{2+} homeostasis.[6,9,12–18,26,27]

Glucose intolerance and insulin resistance seen in diabetes and diabetic-like states (e.g. heart failure, ageing) depress fatty acid desaturase enzyme expression and function, increasing the selective decrease of ω-3 PUFA in membrane phospholipids.[26,27] It has been shown from animal studies that insulin resistance is increased when membrane levels of ω-3 PUFA are low, but can be restored by ω-3 PUFA treatment, thus circumventing the disturbance in fatty acid metabolism due to deficits in delta-5 and delta-6 fatty acid desaturase activities that occur in diabetes.[26,27] In acute type I diabetes in rats, DHA levels in myocardial membranes decline compared with levels of ω-6 PUFA, which remain largely unchanged.[26]

Previous studies with isolated cardiac cells and intact hearts have shown that ω-3 PUFAs specifically normalise the transient outward current, the voltage-dependent Na^+ current, the delayed rectifier current, Ca^{2+}-Mg^{2+}-ATPase, Na^+-Ca^{2+}-exchanger, Ca^{2+} uptake in the sarcoplasmic reticulum and L-type Ca^{2+} current, and prevent Ca^{2+} overload in cytoplasm and mitochondria.[9,13,28,29] They also reduce the activity of membrane phospholipases that are responsible for the generation of key intracellular messengers for Ca^{2+} handling: diacylglycerol and inositol 1,4,5-trisphosphate (IP3). Although considerably more work is required to fully define these and other effects on membrane proteins that contribute to the maintenance of homeostasis, cellular electrical stability, metabolism and sinus rhythm, we expect that restoration of ω-3 PUFA to myocardial membranes confers multiple benefits. Currently, the most convincing clinical benefit relates to a reduction in the vulnerability to the triggers of arrhythmogenesis.

Potential for Therapy

Despite recent controversies in clinical studies, there are considerable clinical data that support the extensive animal data that have demonstrated a specific antiarrhythmic action of dietary fish oil.[14–16] A low incidence of both primary cardiac arrest and post-infarction sudden death plus improved heart rate variability in patients consuming moderate levels of fish or fish oil all strongly indicate that the antiarrhythmic effect of ω-3 PUFA treatment is very potent in humans.[30–35] Studies of post-myocardial infarction patients (and heart failure patients) have identified reduced mortality associated with modest consumption of ω-3 PUFAs (~1g/day) that does not significantly influence classical risk factors such as plasma triacylglycerols, blood pressure or thrombogenic factors, even though these are ameliorated by ω-3 PUFAs at higher doses (~5g/day).[30–35]

Diabetic cardiomyopathy, independent of vascular disease, contributes to high patient morbidity and mortality due to heart failure. Heart failure predisposes to sudden arrhythmic death, and the incidence of heart failure is more than doubled in diabetic patients.[36] Thus, it is not surprising that a fish-rich diet that provides a rich source of ω-3 PUFAs is linked to reduced mortality for all cardiovascular disease.[37]

In 1997, the US Food and Drug Administration (FDA) extensively reviewed over 2,600 studies to conclude that dietary intake of fish oil (DHA+EPA) up to and including 3g per day was safe and had no clinically adverse effects or contraindications.[38] In light of this, as well as recent positive clinical studies, the American Heart Association (AHA) now recommends the use of dietary ω-3 PUFA.[39]

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Notably, a recent population-based cohort study prospectively examined 4,815 elderly adults (≥65 years of age) for 12 years, and found that dietary intake of ω-3 PUFAs from fish was associated with markedly lower incidence of atrial fibrillation (AF).[40] Cardiac surgery represents a predictable acute stress to the heart that precipitates a defined degree of

post-operative AF that is not easily prevented or managed and, if left untreated, may lead to stroke and heart failure. Recently, a study involving pre-operative randomised open-label treatment of elective cardiac surgery patients with only 2g per day/five days of regular ω-3 PUFA capsules (n=79) or placebo (n=81) demonstrated a relative reduction in post-operative AF of 54%. AF in the ω-3 PUFA group was 15.2% and in the placebo group it was 33.3%. Hospital stay was significantly reduced by one full day in the ω-3 group.[41]

Conclusions

With advanced age, ω-3 PUFAs are prone to be diminished in the cardiac membranes of individuals on 'modern' diets common in developed nations. This effect is particularly compounded by disease. ω-3 PUFAs are antiarrhythmic and protective against ischaemia-reperfusion injury, and circumvent the perturbations in cardiac membranes due to advanced age: action potential and contractility, loss of Ca^{2+} homeostasis, inefficient O_2 and energy metabolism, oxidative stress and inflammation. ω-3 PUFA dietary supplementation may target many of these factors, as has been reported by numerous animals studies, and is now required to be better demonstrated in humans.

Large clinical trials have shown that ω-3 PUFA therapy reduces mortality, particularly mortality related to fatal ventricular fibrillation in post-myocardial infarction patients. Early clinical studies suggest that ω-3 PUFA intake via a fish-rich diet is associated with reduced incidence of AF in the elderly general population and after cardiac surgery. However, substantially more work is required to generate the definitive clinical evidence of the therapeutic efficacy of ω-3 PUFA and to address variability in clinical response. In particular, more detailed delineation of all the molecular mechanisms of ω-3 PUFA action in humans is required to determine the extent of its capacity to minimise, circumvent, prevent or reverse key age-linked cardiac membrane dysfunction. The further elucidation of these mechanisms will ultimately generate new therapeutic targets and the potential development of novel therapeutic agents that exert more specific and potent effects. ■

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