A Diet Rich in Unsaturated Fatty Acids Prevents Progression Toward Heart Failure in a Rabbit Model of Pressure and Volume Overload

Hester M. Den Ruijter, PhD; Arie O. Verkerk, PhD; Cees A. Schumacher, RA; Sander M. Houten, PhD; Charly N.W. Belterman, RA; Antonius Baartscheer, PhD; Ingeborg A. Brouwer, PhD; Marc van Bilsen, PhD; Baukje de Roos, PhD; Ruben Coronel, MD, PhD

Background—During heart failure (HF), cardiac metabolic substrate preference changes from fatty acid (FA) toward glucose oxidation. This change may cause progression toward heart failure. We hypothesize that a diet rich in FAs may prevent this process, and that dietary ω-3 FAs have an added antiarrhythmic effect based on action potential (AP) shortening in animals with HF.

Methods and Results—Rabbits were fed a diet containing 1.25% (w/w) high oleic sunflower oil (HF-ω9, N=11), 1.25% fish oil (HF-ω3, N=11), or no supplement (HF-control, N=8). Subsequently, HF was induced by volume and pressure overload. After 4 months, HF-parameters were assessed, electrocardiograms were recorded, and blood and ventricular tissue were collected. Myocytes were isolated for patch clamp or intracellular Ca2+ recordings to study electrophysiologic remodeling and arrhythmogenesis. Both the HF-ω9 and the HF-ω3 groups had larger myocardial FA oxidation capacity than HF control. The HF-ω3 group had significantly lower mean (± SEM) relative heart and lung weight (3.3±0.13 and 3.2±0.12 g kg⁻¹, respectively) than HF control (4.8±0.30 and 4.5±0.23), and shorter QTc intervals (167±2.6 versus 182±6.4). The HF-ω9 also displayed a significantly reduced relative heart weight (3.6±0.26), but had similar QTc (179±4.3) compared with HF control. AP duration in the HF-ω3 group was ~20% shorter due to increased I_{to} and I_{K1} and triggered activity, and Ca2+-aftertransients were less than in the HF-ω9 group.

Conclusions—Dietary unsaturated FAs started prior to induction of HF prevent hypertrophy and HF. In addition, fish oil FAs prevent HF-induced electrophysiologic remodeling and arrhythmias. (Circ Heart Fail. 2012;5:376-384.)

Key Words: heart failure ■ nutrition ■ lipids ■ remodeling heart failure ■ electrophysiology

In heart failure (HF), metabolic substrate preference shifts from fatty acid oxidation (FAO) toward glucose oxidation. It has been suggested that this change underlies the progression toward HF. As a consequence, driving the balance toward FAO is expected to prevent or slow down the progression of HF and reduce the associated arrhythmias. We hypothesize that this can be accomplished by a diet rich in fatty acids (FAs).

Clinical Perspective on p 384

Heart failure is associated with a prolongation of ventricular action potential (AP), which plays an important role in the genesis of life-threatening ventricular tachyarrhythmias. Dietary ω-3 FAs cause a shortening of the AP duration (APD) in healthy animals. Acute application of ω-3 FAs to isolated myocytes from hearts of patients with HF led to APD shortening and reduced incidence of triggered activity. It is not known whether dietary ω-3 FAs exert the same action in vivo.

The Diet and Reinfarction and GISSI trials (Gruppo Italiano per lo Studio della Sopravvivenza nell’Infarto miocardico) addressed the potential beneficial effects of the Mediterranean diet and showed that increased intake of fish oil reduced cardiovascular death following a myocardial infarction. This mainly was attributed to risk reductions in sudden death in the GISSI Trial. Because ventricular tachyarrhythmias often precede sudden death, the effect of fish oil on arrhythmogenesis has been studied intensively in...
unsaturated (in the first but not the second GISSI trial.11 Absolute risk reduction in total mortality compared with the original GISSI trial.11 The difference between the 2 GISSI trials7,11 is that in the latter fish oil was provided to patients that had already developed severe HF,11 whereas in the first GISSI trial, patients with a recent myocardial infarction and with developing hypertrophy were included.7 The different outcomes between trials may be explained by a diet-induced retardation of the progression of HF in the first but not the second GISSI trial.

We tested whether long term supplementation of the diet with unsaturated (ω3- or ω9-) FAs prior to the development of HF by combined volume and pressure overload in rabbits attenuates structural and functional remodeling and leads to less hypertrophy and arrhythmias. Rabbits without dietary supplements were used as control. We measured parameters of HF, electrocardiograms, and markers of FAO, as well as triggered arrhythmias and cardiac electrophysiological parameters 4 months after the induction of HF.

Methods

Design of the Study

The investigation was approved by the local ethical committee and conformed to the Guide for the Care and Use of Laboratory Animals (NIH Publication No. 85–23, revised 1996).

The design of the study is shown in Figure 1. Male New Zealand White rabbits (2.5–3 kg) were allocated to 1 of 4 groups. Group 1 consisted of rabbits on a diet (150 g/d) without added FAs that underwent volume and pressure overload (HF control; n=8). Group 2 and 3 consisted of rabbits on a diet (150 g/d) supplemented with either 1.25% (w/w) ω9-sunflower oil (HF-ω9; n=11) or ω3-fish oil (HF-ω3; n=11) that underwent volume and pressure overload. Group 4 consisted of rabbits on a diet (150 g/d) without added FAs (control; n=5).

Heart failure was induced by combined volume and pressure overload as described before, typically inducing concentric hypertrophy as well as some left ventricular dilatation of the heart.2,12 Volume overload was induced by aortic valve rupture (100% pulse pressure increase). Three weeks later, pressure overload was created by a ≤50% suprarenal aortic stenosis. After 4 months, the rabbits were anesthetized by a combination of ketamine (50 mg im) and xylazine (10 mg im), blood was collected, and an ECG was recorded. The rabbits were then heparinized (5000 IU) and killed by intravenous injection of pentobarbital (240 mg). Body weight, heart weight, and lung weight were measured, and the presence of ascites and pericardial and thoracic fluid documented.10 Left ventricular tissue was collected and stored at −80°C.

Diets

The diets (RDS, Wijk bij Duurstede, NL) contained ~3.5% (w/w) fat, mainly linoleic acid. The addition of 1.25% sunflower oil and 1.25% ω3-fish oil increased mono-unsaturated FAs in the ω9-diet (C18:1ω9), and eicosapentaenoic acid and docosahexaenoic acid (EPA-C20:5ω3 and DHA-C22:6ω3) in the ω3-diet (see online-only Data Supplement Table 1).

Analysis of Diet, Tissue, and Plasma

Lipids from the food and tissue were extracted according to Folch et al. Phospholipids were isolated with Bond Elut Aminopropyl solid phase extraction columns. Saponification and methylation of the phospholipids with boron trifluoride (Pierce) was performed, and the formed FA methyl esters were subjected to capillary gas chromatography using a Chrompack column (Fused Silica, Chrompack), a flame ionization detector, and H2 as carrier gas. FA methyl esters were expressed as a fraction of the total amount. To determine the plasma lipid profiles (total, low density lipoprotein [LDL], and high density lipoprotein [HDL] cholesterol, and triglycerides), samples were analyzed with spectrophotometry (enzymatic color test) using commercial kits (Roche Diagnostics GmbH).

Cell Preparation

Midmyocardial myocytes were isolated by enzymatic dissociation from the left ventricular free wall.14 Aliquots of cells were superfused (37°C) in a recording chamber on the stage of an inverted microscope. Quiescent rod-shaped cross-striated cells with a smooth surface were selected.

Electrophysiology

Electrocardiogram

QT segments were corrected for heart rate using linear regression analysis obtained from 37 healthy control rabbits. Accordingly, the QT intervals were normalized to an RR interval of 4 Hz using the formula QTc=QT−0.26*(RR−250).

Current-Clamp and Voltage-Clamp

See the online-only Data Supplement.

Ca2+ Transients

Intracellular Ca2+ (Ca2++) was measured in indo-1-am loaded myocytes as described previously.12 Dual wavelength emission of indo-1 was recorded (405–440/505–540 nm, excitation at 340 nm), and free Ca2++, was calculated.12

Gene Expression and Enzymatic Activity Determination

Left ventricular samples were homogenized and total RNA was isolated. An additional wash step of 70% ethanol increased the RNA purity. Integrity of the RNA was checked by means of 260/280 ratio. cDNA synthesis and quantitative polymerase chain reaction (qPCR) assays were performed (for primers see online-only Data Supplement Table II).15 Results were normalized to the geometric mean of 2 reference genes, that is, cyclophilin A and hypoxanthine guanine phosphoribosyltransferase,16 using qBase software.17

Activity

Left ventricular tissue was homogenized in cold phosphate-buffered saline followed by sonication on ice (twice 40J at ~8W output). Homogenates were centrifuged for 10 minutes at 1000 g. Carnitine palmitoyltransferase 1 (CPT 1) activity was assayed by measuring the [U-13C] palmitoylcarnitine from carnitine, and [U-13C] palmitoyl-CoA on the tandem-mass spectrometer as previously described.18 Incubations were performed in the presence and absence of 0.2 mmol/L malonyl-CoA. The activity inhibited by malonyl-CoA was considered to represent CPT 1.
Proteomics
Cardiac tissue protein homogenates were prepared by grinding frozen tissue from 5 HF control, 8 HF-ω9, and 10 HF-ω3 rabbits in liquid nitrogen and homogenizing them in a lysis buffer (7 mol/L Urea, 2 mol/L Thiourea, 2% CHAPS [3-[(3-cholamidopropyl)dimethylammonio]-1-propane-sulfonic acid], 0.06% protease inhibitor cocktail [Roche]). Protein concentrations were determined using the RC/DC [reducing agent compatible, as well as detergent compatible] assay (BioRad), and 300 μg of protein were loaded per gel. BioRad IPG strips (pI 5–8) were used for the separation of proteins in the first dimension, and SDS-PAGE was performed on 18x18 cm acrylamide gradient (8%–16%) gels for the separation of proteins in the second dimension. Flamingo (Biorad) fluorescent-stained gels were analyzed using SameSpots (Nonlinear Dynamics). Spot density values were exported from the SameSpots software into R 2.11.1 (R Foundation for Statistical Computing) to perform statistical analysis as described below. Spots were excised from the gel using a robotic spot cutter (BioRad), trypsinized using a MassPrep Station (Micromass), and analyzed by LC-ESI-MS/MS.

Statistics
Data are mean±SEM (n,N: number of cells, rabbits). Data were statistically analyzed using 1- or 2-way ANOVA, on ranks (with Kruskal-Wallis test) or repeated measures ANOVA if appropriate. In repeated measures ANOVA, voltage steps were modeled as a categorical factor and cells as the repeated factor, without interaction. Post hoc testing for multiple comparisons was done with the Holm-Sidak, Fisher LSD Method (with parametric ANOVA), or Dunn test (with nonparametric ANOVA); P<0.05 was considered statistically significant.

Results
Fatty Acid Oxidation
Left ventricular activity of CPT 1, a rate-limiting enzyme controlling mitochondrial import of FAs,20 was increased significantly in the HF-ω9 and HF-ω3 groups compared with HF control and control (Figure 2A). The cardiac mRNA level of the α-subunit of mitochondrial trifunctional protein (HADHA), another FAO enzyme, was markedly decreased in the HF control group compared with healthy control rabbits. The ω9- and ω3-FA-enriched diets prevented the HF-mediated decline in HADHA mRNA levels and even resulted in significantly higher levels than in healthy control rabbits (Figure 2B). There were no statistical differences in fat composition between the HF-ω9 and the HF-control groups.

Plasma and Tissue Content
The diets rich in ω3-FAs resulted in a significant reduction of plasma triglycerides compared with HF control and HF-ω9. Triglyceride content in the ω3-group was not different from that of control rabbits (Table). There were no differences in total cholesterol or LDL- or HDL-cholesterol between the groups (data not shown).

In the HF-ω3 group, the ω3-FA tissue-content comprised 15.5±0.5% of the total FAs extracted compared with 5.3±0.3% in the HF control group and 4.7±0.2% in the HF-ω9 group. The incorporation of ω3-FAs was at the expense of ω6-FAs and ω9-FAs. There were no statistical differences in fat composition in the heart between the HF-ω9 and the HF-control groups. Supplemental Table III shows the myocardial phospholipid fatty acid composition in the 4 groups (see online-only Data Supplement). The percentage of ω9-FA oleic acid in the myocardial phospholipids is highest in the HF-ω9 group and in the control group, and lowest in the HF-ω3 group.

Unsaturated Fatty Acids Prevent Cardiac Hypertrophy During Developing Heart Failure
Weight gain was similar in the 4 groups (Table). The increase of the aortic pulse pressure immediately after aortic valve
rupture was 102 ± 1.3, 101 ± 0.9, and 100 ± 0.7% for HF control, HF-ω9, and HF-ω3, respectively (P > 0.05).

The HF control group had a significantly larger (relative) heart and lung weight than the control rabbits. In addition, the majority of the HF control rabbits had ascites and thoracic and pericardial fluid (Table). Although the trigger for the development of hypertrophy and HF was the same in all HF groups, the animals that received diets rich in ω9- and ω3-FAs had a significantly reduced relative heart weight compared with HF control (P < 0.05, ANOVA followed by Fisher least significant difference (LSD) test; Table). In addition, relative lung weight was lower in the HF-ω3 group than HF control and did not differ from the control rabbits (P < 0.05, ANOVA followed by Fisher LSD test; Table). Supplemental Table IV shows the echocardiographic data that reflect the heart weight data (see online-only Data Supplement).

Pro-brain natriuretic peptide mRNA levels were measured as an indicator of cardiac hypertrophy. Relative myocardial mRNA levels of BNP were significantly lower in the HF-ω9 and HF-ω3 groups than the control rabbits, but this was significantly less than the HF control group.

QTC was shorter in the HF-ω3 rabbits than HF-control and HF-ω9, and similar to that of healthy controls (Table). The heart rate did not differ between the groups. Figure 3A displays typical APs recorded from isolated myocytes of the 4 groups at 1 Hz pacing frequency. AP duration at 90% repolarization (APD90) is prolonged significantly in the HF control group compared with control at pacing frequencies below 3 Hz (Figure 3B). Whereas APD90 in the HF-ω9 group did not differ from HF-control, it was significantly shorter in the HF-ω3 group than in the HF control group and not different from control (Figure 3B). Similarly, the AP at 50% repolarization (APD50) was 202 ± 8 and 197 ± 16 ms, respectively) than control (174 ± 6 ms). The APD50 in the HF-ω3 group (172 ± 10 ms) was similar to that of healthy control rabbits. Both HF-ω3 and control were significantly different from HF control and HF-ω9 (all P < 0.05, repeated measures ANOVA). Cell capacitance data reflected heart weight (see online-only Data Supplement Table V).

Figure 3C shows representative examples of Ca2+ transients in 2 Hz stimulated myocytes of all groups. Myocytes of the HF-control group have increased diastolic [Ca2+]i, decreased Ca2+ transient amplitude, and slowed relaxation compared with control and in HF-ω3 groups. In the HF-ω9 group, diastolic Ca2+ was not different from the HF group, and thus increased compared with control. In the same HF-ω9 group, however, Ca2+ transient amplitude and relaxation time were not different compared with control and HF-ω3 groups. Data are summarized in Figure 3D. Diastolic Ca2+ levels were significantly lower in the HF-ω3 (98 ± 7.9 nmol/L, n = 19, P < 0.05 ANOVA followed by Holm-Sidak test) than in HF control (128 ± 8.4 nmol/L, n = 26) and HF-ω9 (120 ± 6.8
Dietary Fish Oil Increases \( I_{\text{to}} \) and \( I_{\text{K,1}} \) in Heart Failure

To investigate the mechanism by which dietary fish oil prevents AP prolongation, we studied the major ion currents in the 4 groups (Figure 4A).

**L-Type Ca\(^{2+}\) Current**

Figure 4B shows typical examples and current-voltage (I-V) relationships of the L-type Ca\(^{2+}\) current (\( I_{\text{Ca,L}} \)). Mean \( I_{\text{Ca,L}} \) densities were similar in the control, HF control, and HF-\( \omega \)3 group (Figure 4B). In the HF-\( \omega \)9 group, \( I_{\text{Ca,L}} \) was significantly larger at plateau potentials (–5 to +5 mV) compared with all other groups (P<0.05 repeated measures ANOVA). Voltage-dependency of \( I_{\text{Ca,L}} \) (in)activation did not differ significantly between the 4 groups (data not shown).

**Na\(^{+}\)-Ca\(^{2+}\) Exchange Current**

Figure 4C shows typical examples and I-V relationships of the Na\(^{+}\)-Ca\(^{2+}\) exchange current (\( I_{\text{NCX}} \)). There were no differences in the reverse (outward) mode and in the forward (inward) mode of the \( I_{\text{NCX}} \) between the 4 groups (Figure 4C).

**K\(^{+}\) Currents**

Figure 5A shows typical examples and I-V relationships of the transient outward K\(^{+}\) current (\( I_{\text{to}} \)). Mean \( I_{\text{to}} \) densities were significantly larger in the HF-\( \omega \)3 group than HF control (\( P<0.05 \) at +20 and +30 mV, repeated measures ANOVA; Figure 5A). In the HF-\( \omega \)9 group, \( I_{\text{to}} \) was larger at +30 mV than HF control. Voltage dependency of \( I_{\text{to}} \) activation and inactivation did not differ significantly between the 4 groups (data not shown).

**Fish Oil Inhibits Triggered Arrhythmias**

Representative examples of transmembrane potentials in the 4 groups in the presence of 1 \( \mu \)mol/L noradrenalin after rapid pacing are shown in Figure 6A. In the HF control and the HF-\( \omega \)9 myocytes, the last stimulated AP (arrow) is followed by delayed after depolarizations (DADs) and a triggered AP. In the HF-\( \omega \)3 myocyte, a DAD was present, but triggered APs did not occur. In isolated myocyte of the control group no arrhythmias occurred. Triggered APs and DADs were abundantly present in the HF control and in the HF-\( \omega \)9 group compared with control (both \( P<0.05 \), nonparametric ANOVA followed by Dunn test; Figure 6B). There was a significant reduction in both triggered APs and DADs in the HF-\( \omega \)3 group compared with control."
The incidence of triggered APs was 17% and 14% in the HF control and HF-α9 group, respectively, and significantly less in the control (0%) and HF-α3 myocytes (7%).

**Proteomics**

A hypothesis-free proteomic approach was followed to identify potential protein expression changes that could underlie the electrophysiological differences between the

---

**Figure 4.**

A, Schematic representation of the action potential and its major ion currents. B, Top: 2-step protocol for L-type Ca²⁺ current (\(I_{Ca-L}\)) measurement. Middle: typical examples of \(I_{Ca-L}\) during voltage steps to 0 mV. Bottom: average current-voltage (I-V) relationship of \(I_{Ca-L}\). *\(P<0.05\) versus HF control; †\(P<0.05\) versus control; ‡\(P<0.05\) versus HF-α3 (repeated measures ANOVA). C, Top: the Na⁺-Ca²⁺ exchange current (\(I_{NCX}\)) was measured in Ca²⁺-buffered conditions as the Ni²⁺-sensitive current during a descending voltage ramp protocol. Middle: typical examples of \(I_{NCX}\). Bottom: average I-V relationship of \(I_{NCX}\).

**Figure 5.**

A, Top: 2-step protocol for transient outward K⁺ current (\(I_{to1}\)) measurement. Middle: typical examples of \(I_{to1}\) during voltage steps to 40 mV. Bottom: average current-voltage (I-V) relationship of \(I_{to1}\). *\(P<0.05\) versus HF control; †\(P<0.05\) versus control; ‡\(P<0.05\) versus HF-α3 (repeated measures ANOVA). C, Top: protocol for delayed rectifier K⁺ current (\(I_{Kr}\)) measurement. Middle: typical examples of \(I_{Kr}\) during voltage steps to 10 mV. \(I_{Kr}\) was defined as the tail current on stepping back to the holding potential. Bottom: average I-V relationship of \(I_{Kr}\).
However, the relatively long action potential, and increased makes the heart less vulnerable for re-entrant arrhythmias. Thus, arrhythmogenic triggers as in the HF control group. Thus, reduce both hypertrophy and triggered arrhythmias. The data support the notion that unsaturated (fish oil) FAs suppress cardiomyocyte hypertrophy in vitro and in vivo, and we speculate that this was affected by activating proliferator-activated receptors.

The cardioprotective mechanisms of fish oils are diverse, and include antiarrhythmic, antiatherosclerotic, and anti-inflammatory effects. In addition, ω3-FAs incorporate in the mitochondrial membranes of cardiac myocytes and change FAO and mitochondrial respiration.

### Omega-3 and Omega-9 Fatty Acids Prevent Cardiac Hypertrophy

We found that both dietary ω3- and ω9-FAs prevented cardiac hypertrophy following combined volume and pressure overload. Epidemiological studies show that increased intake of ω3-FAs is associated with reduced incidence of HF. Similarly, fish oil diets in rats with pressure overload also prevented hypertrophy. Although there is limited experimental and clinical evidence of the role of high fat diets in the pathogenesis of HF, both animal studies and human studies show that high unsaturated fat diets are not harmful for the heart and may even improve biomarkers of cardiovascular disease.

### Omega-3 Fatty Acids Are Increased in Cardiac Tissue

In the HF-ω3 group, this diet increased myocardial levels of ω3-FAs by ≈10% at the expense of ω6-FAs. In patients, cardiac ω3-FAs may increase up to ≈6% after fish oil supplementation for 1 month. The similarity between our data and the levels obtained in man suggests that the effects observed in this study are relevant in human HF.

Note that the vaccenic acid (18:1n7) percentage is low, but highest in the HF-ω3 group, suggesting that this trans FA did not have any detrimental effect in our study, as suggested recently.

### Unsaturated Fatty Acids Lower Triglycerides During Developing Heart Failure

Our data show that the HF-ω3 and HF-ω9 groups both had lower plasma triglycerides levels than the HF control group. Omega-3 FAs appear to be superior in lowering plasma triglycerides, as these were lower in the HF-ω3 group than in the HF-ω9 group. Also in humans, the Mediterranean diet, rich in monounsaturated fatty acids primarily from olives and olive oil, and fish oils markedly decreased triglyceride levels. Therefore, monounsaturated fatty acids not only have antihypertrophic effects, but also have favorable metabolic effects.
Dietary Fish Oil But Not Sunflower Oil Prevents Electric Remodeling in Heart Failure

In addition to a slowing effect on the progression of HF, the fish oil diet resulted in a reduced QTc interval on the surface ECG. This observation was supported in isolated myocytes of the HF-ω3 group that displayed shorter APs. In all cardiomyopathy models, Ito1 density is reduced. This was confirmed in our study in which we observed a ≈50% decrease in Ito in the HF control group. The HF-ω3 and the HF-ω9 group did not display remodeling of Ito, which explains the shorter AP in the HF-ω3 group. In the HF-ω9 group, however, ICa,L was larger than in the other groups, explaining the longer APs and the higher diastolic Ca2+ levels compared with the HF-ω3 group. In addition, increased Ca2+ influx through L-type Ca2+ channels by necessity leads, under steady state conditions, to increased efflux of Ca2+ through the Na+/Ca2+ exchanger and to increased depolarizing ICaX and AP prolongation. Because calcium handling was impaired in the HF-ω9 group despite the normalization of SERCA2a mRNA, we cannot exclude that sarco/endoplasmatic reticulum calcium ATPase function was compromised nevertheless.

Fish oil has disparate effects on ion currents, and both an increase and decrease in K+ currents have been described. This appeared to depend on whether the FAs were applied to the cells or incorporated in the membranes. Our study supports the idea that the effect of fish oil on cardiac arrhythmias not only depends on the mode of application, but also on the underlying cardiac pathology.

We followed a proteomics approach to identify potential protein expression changes between the HF-ω9, HF-ω3, and control groups. This failed to show major significant differences between groups, apart from an indication that oxidation of very long chain FAs and oxidative phosphorylation may be increased in HF-ω9, and to a lesser extent HF-ω3. Therefore, the observed electrophysiological differences between the HF-ω9 and HF-ω3 groups cannot be explained by regulation of visualized proteins in the 2D gel, albeit that our approach may not have been sufficiently sensitive to detect regulation of pore forming and accessory subunits for each individual ion channel that may have been affected by the dietary interventions.

Potential Significance and Limitations

Dietary fish oil supplements are recommended for patients after a myocardial infarction, as their cardioprotective efficacy has been demonstrated in various clinical trials on the Mediterranean diet. The result of the recent GISSI-HF, however, showed a modest protection against cardiovascular death and hospitalization in patients with established HF.

We studied the effect of fish oil during the development of HF without interference of medical therapies. In this setting, fish oil supplementation prevented both structural and electric remodeling associated with HF. This may explain why fish oil supplementation causes a larger reduction in sudden death in patients at risk for HF (after acute myocardial infarction) than in patients with established HF, even in the presence of optimal medical therapy. Indeed, increased fish oil consumption is reported to be associated with a reduced incidence of HF.

We would argue that the traditional diet-heart hypothesis may need to be adjusted after confirmation of our findings in man. The molecular mechanisms underlying these effects remain uncertain, although our data suggest that an increased capacity for myocardial FAO plays a role, as well as the alteration of ion channel kinetics.

We were unable to specify the phospholipid content of the sarcolemma in the 4 groups, and were limited by the paucity of suitable antibodies for validation of protein changes.

Conclusion

Dietary unsaturated FAs supplementation initiated before the development of HF reduces hypertrophy. In addition, fish oil FAs prevent HF-induced electric remodeling and triggered arrhythmias. Our study may explain why fish oil FAs are powerful antiarrhythmic drugs during the development of HF.

Acknowledgments

The authors thank Betty van der Struijs, Chantal Munts, and Jan Ruijter for excellent support.

Sources of Funding

This work was supported by the Netherlands Heart Foundation (grants 2003B079 and 2007B018), the SEAFOODplus program of the European Union (grant 506359), EU FP6 grant LSHM-CT-2005-018833/EUGeneHeart to M.v.B., and the Netherlands Organisation for Scientific Research (VIDI-grant No. 016.086.336 to S.M.H.).

Disclosures

None.

References


20. Duda MK. O’Shea KM, Tintinu A, Xu W, Khairallah RJ, Barrows BR, Chess DJ, Azimzadeh AM, Harris WS, Sharov VG, Sabbath HN, Stanley WC. Fish oil-fed animals and was caused by increased Ito1 and IK1, and resulted in reduced triggered arrhythmias in the fish oil-fed group. *Circulation.* 2008;118:1222–1228.


27. Duda MK, O’Shea KM, Tintinu A, Xu W, Khairallah RJ, Barrows BR, Chess DJ, Azimzadeh AM, Harris WS, Sharov VG, Sabbath HN, Stanley WC. Fish oil-fed animals and was caused by increased Ito1 and IK1, and resulted in reduced triggered arrhythmias in the fish oil-fed group. *Circulation.* 2008;118:1222–1228.


---

**CLINICAL PERSPECTIVE**

During heart failure, the cardiac metabolic substrate preference changes from fatty acid (FA) toward glucose. This change may contribute to progression of disease. We addressed whether a diet rich in unsaturated fatty acids limits progression toward HF. We fed rabbits with a diet rich in sunflower oil, fish oil, or control feed, and HF was induced by volume and pressure overload. Both the FA-supplemented groups had increased myocardial FA oxidation. The sunflower oil group had lower relative heart weight and had similar QTc interval as the HF control group. The fish oil group also displayed lower heart weight, but had shorter QTc intervals on the ECG. The shorter QTc was explained by action potential shortening in fish oil-fed animals and was caused by increased Ito1 and IK1, and resulted in reduced triggered arrhythmias in the fish oil group. Thus, dietary supplementation with unsaturated fatty acids prevents hypertrophy and HF, and fish oil supplementation additionally prevents HF-induced electric remodeling. These data may contribute to understanding the potential cardioprotective effects of Mediterranean diets in this setting. In addition, our data may shed light on why fish oil supplementation is beneficial in patients with a recent myocardial infarction, but not in patients with established HF.