Lecture 4a: Mitochondrial involvement in cardiovascular disease

Cardiovascular disease mortality – surely a success story?

- For years, CVD mortality has been declining.
- However, the **rate of decline in CVD mortality has slowed considerably in recent years** in most developed countries for both males and females, particularly at ages 35–74 years.
- **CVD mortality is now rising in 12 out of the 23 nations** studied in 2017.
- The researchers attribute this to increasing obesity and health inequalities, especially in English-speaking nations.

(Lopez AD, Int J Epidemiol, 2019)

Mitochondria and heart function

- The heart depends on mitochondria for 95% of its ATP, the remaining 5% coming from glycolysis. **The heart has one of the highest energy demands in the human body.** Every day, cardiac mitochondria have to synthesise 6kg of ATP to meet cardiac energy requirements for contraction. An adult cardiac myocyte contains roughly 6000 mitochondria, constituting up to 40% of cell volume.
- **The heart also requires large and constant supplies of oxygenated blood to enable OXPHOS.**
- This makes the cardiovascular system **uniquely vulnerable** and it is likely that everyone with cardiovascular disease (CVD) will have some form of **energy deficiency.**
- Furthermore, **cardiac myocyte mitochondria cannot travel within the myocyte and they do not undergo fission or fusion**, putting them at a **disadvantage to the mitochondria in other tissues**.

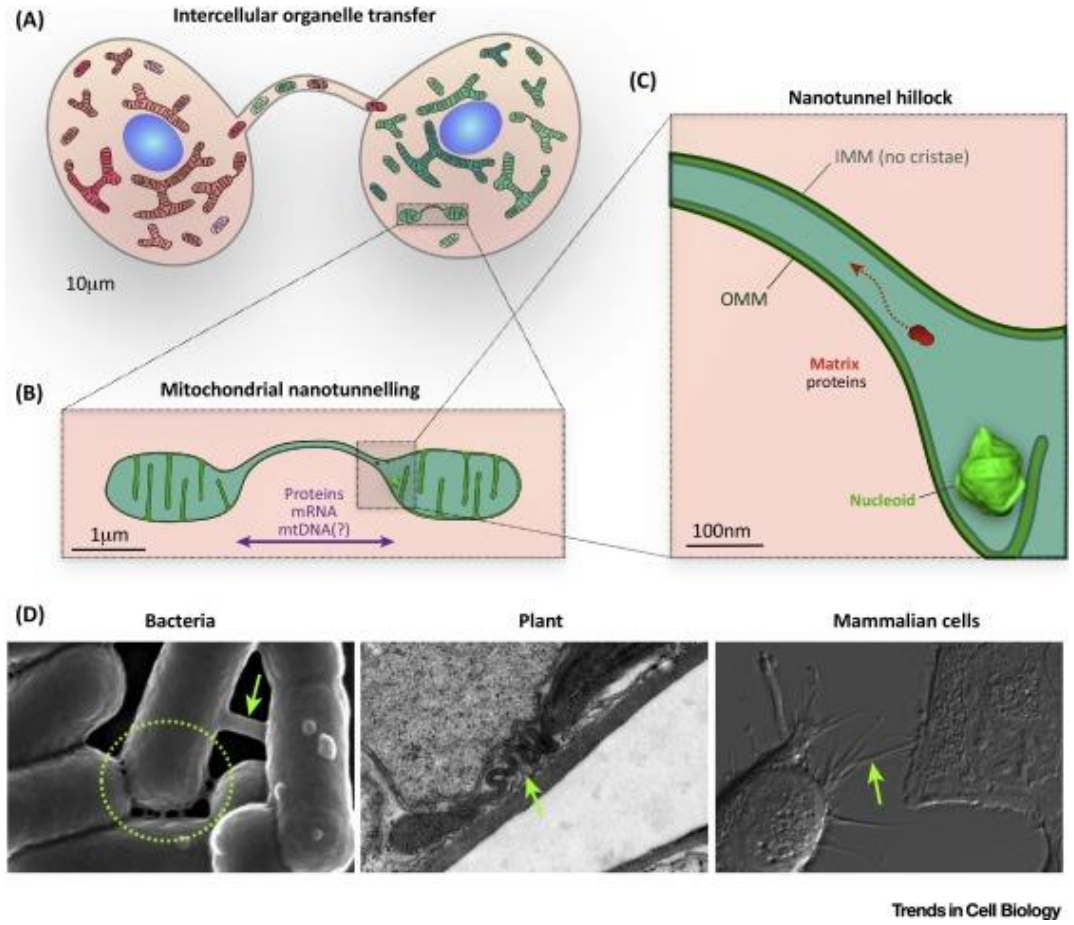
(Sheeran FL, Adv Exp Med Biol, 2017; Huang X, Proc Natl Acad Sci, 2013; Gustafsson AB, Cardiovasc Res, 2008)

How cardiac myocytes overcome their limitations

- Cardiac myocytes have evolved an ingenious mean of circumventing these limitations, not seen in other body systems: they form one **large static interconnected network of mitochondria in each cell.**
- By this means, they **co-ordinate ATP production and exchange parts to repair damaged mitochondria.**
- They achieve this by:
  - **adjacent mitochondria 'kissing';**
  - **building a nanotunnel between nonadjacent mitochondria**.

(Huang X, Proc Natl Acad Sci, 2013; Lou E, PLoS One, 2012)

Pictures of mitochondrial nanotunnels and kissing



Vincent AE, Trends Cell Biol, 2017

Mitochondria in CVD development and progression

- **Angina, hypertension, atherosclerosis, ischaemia, heart failure and diastolic dysfunction all have their roots in failure of mitochondrial energy production**. Any degree of ischaemia will worsen pre-existing conditions by reducing available ATP.
- **In the heart, the preferred method of producing ATP is β-oxidation of fatty acids or ketones.** Pyruvate contributes minimally to ATP synthesis in the healthy heart. **As the heart shifts to the less efficient glycolysis for energy production, lactic acid build-up develops**, with a progressive loss of contractility and risk of angina pain.
- **This incomplete β-oxidation in the heart has been associated with heart failure, ischaemic heart disease and diabetic cardiomyopathy,** producing an accumulation of lipid peroxides.
- **Heart failure is associated with a gradual decline in the bioenergetic reserve capacity of the myocardium and decreased mitochondrial biogenesis.**
- In addition, mitochondrial dysfunction is involved in every aspect of atherosclerosis development and progression. Atherosclerosis-associated inflammation is linked to mitochondrial dysfunction and mitochondria themselves can act as pro-inflammatory agents, leading some to suggest atherosclerosis may be an autoimmune disease.

(Sheeran FL, Adv Exp Med Biol, 2017; Jiang Y, Toxicology, 2015; Silva-Oropeza E, 2004; Bonora M, Nat Rev, 2019; Montaigne D, Card Electrophysiol Clin, 2015; Lee TW, J Biomed Sci. 2017; Botto N, Mutat Res, 2005; Bray AW, Curr Opin Cardiol, 2017; Komlodi T, J Bioenerg Biomembr, 2018; Robb EL, J Biol Chem, 2018)

Implications of the loss of mitochondrial energy production

- Adenine nucleotide (ATP, ADP, AMP) levels decline more than 30% in heart failure and more than 40% in coronary artery disease and ischaemic heart disease. Tissue biopsy and nuclear magnetic resonance findings confirm these observations.
- High concentrations of ATP are required to activate calcium pumps necessary to facilitate cardiac relaxation and diastolic filling. More ATP is needed to fill the heart (diastole) than to empty it (systole).
- Low ATP levels disable calcium channels in the heart, so that ions cannot escape during the ejection phase, preventing the heart muscle from relaxing (i.e. diastolic dysfunction) and build up of excess calcium ions.
- In ischaemic or hypoxic hearts, the cell's ability to match ATP supply and demand is disrupted, leading to both depletion of the cardiac energy pool and dysfunction in mitochondrial turnover mechanisms. When ATP levels fall, diastolic heart function deteriorates; diastolic dysfunction is an early sign of myocardial failure despite the presence of normal systolic function and preserved ejection fraction.
- The heart tries to compensate by enlarging its size (hypertrophy) but this worsens the ejection fraction and diastolic function.
- Reverse electron transport is a key source of mtROS production, especially in ischaemia-reperfusion injury.
- The preservation of ATP is vital in the heart, especially during bouts of ischaemia, as a reduction in ATP level corresponds to a loss of diastolic function. The larger the energy reserves during ischaemia, the greater the chance that ATP recovery can occur upon reperfusion and the lower the extent of cardiac injury.
- Several mutations in genes coding for components of the mitochondrial respiratory chain have been associated with familial cardiomyopathies; heart tissue from patients with CAD contains many more mtDNA mutations compared to age-matched controls.

(Sinatra ST, Alternative Therapies, 2009; Ingwall JS, Circ Res, 2004; Bashore TM, JACC, 1987; Mirogoli M, Cell Rep, 2016; Santulli G, Proc Natl Acad Sci USA, 2015; Bonora M, Nat Rev, 2019; Glanz VY, Front Biosci, 2020; Rosca MG, Heart Fail Rev, 2013; Sheeran FL, Adv Exp Med Biol, 2017; Jiang Y, Toxicology, 2015; Silva-Oropeza E, 2004; Montaigne D, Card Electrophysiol Clin, 2015; Lee TW, J Biomed Sci. 2017)

Mitochondria and intracellular calcium in heart disease

- Calcium fluxes are at the core of overall cardiac activity. Therefore, defects in the capacity of the mitochondrial network (in conjunction with the endoplasmic reticulum) to regulate calcium homeostasis can alter cardiac functions such as electrical conduction.
- In particular, cardiac cells have a potential to accumulate calcium. Low ATP levels disable calcium channels in the heart, so that ions cannot escape during the ejection phase, preventing the heart muscle from relaxing (i.e. diastolic dysfunction).
- Arrhythmogenesis due to calcium propagation during heart failure is a major clinical problem. Heart failure progressively stiffens the membrane and disrupts the microtubules allowing mitochondrial network communication, displaces mitochondria and causes calcium release. Uncoupling the mitochondrial proton gradient abolished abnormal calcium propagation in heart failure.
- Diastolic calcium leak from the (sarco)endoplasmic reticulum, caused mitochondrial calcium overload and dysfunction in a mouse model of post-myocardial infarction heart failure.

(Mirogoli M, Cell Rep, 2016; Santulli G, Proc Natl Acad Sci USA, 2015; Bonora M, Nat Rev, 2019)

Atherosclerosis and mitochondria

- Mitochondrial dysfunction is involved in every aspect of atherosclerosis development and progression.
- In particular, atherosclerosis-associated inflammation is linked to mitochondrial dysfunction; mitochondria not only trigger the response to external signals, but also can act as pro-inflammatory agents themselves. In this regard, Glanz et al consider that atherosclerosis is potentially an autoimmune disease.
- Damaged mitochondria accumulating in the cytosol of cardiomyocytes or endothelial cells can drive pathogenic inflammatory responses leading to heart disease or atherosclerosis.

(Glanz VY, Front Biosci, 2020; Bonora M, Nat Rev, 2019)

Cholesterol and mitochondria

- Cholesterol is essential for the survival of all cells and for the structure, fluidity and integrity of cell membranes and intracellular lipids. Steroid hormones, bile acids and vitamin D are all dependent on cholesterol for their synthesis. In the elderly, lower cholesterol increased mortality, rather than reducing it.
- Cholesterol is produced in the mitochondria and endoplasmic reticulum through the action of enzymes, principally HMG-CoA reductase, which also synthesises CoQ10.
- But insulin resistance (and other risk factors) can cause HMG-CoA reductase to work overtime producing cholesterol, suggesting that high levels of cholesterol is not a pathology in itself but is merely a response to other pathology.

(Schatz U, Lancet, 2001; Ferramosca A, Adv Nutr, 2014)

Hypertension and mitochondria

- Mitochondria play an essential role in the regulation of vascular smooth muscle cell calcium signalling and buffer excess calcium. Cross-talk between signalling systems regulates vascular tone in response to complex stimulation to maintain blood pressure.
- Vasoconstriction is mediated by angiotensin II and endothelin through activating NADPH oxidase, which produces superoxide to antagonise endothelial nitric oxide synthase (eNOS) in the vascular endothelium. Hence, healthy blood pressure is maintained by the opposing action of 2 free radicals: superoxide and nitric oxide.
- Various stressors such as insulin resistance can reduce eNOS, while mitochondrial biopterin can activate eNOS. Loss of mitochondrial biopterin can lead to increased superoxide production and reduced antioxidants in the vascular endothelium, resulting in hypertension.
- However, shear stress (increased blood flow putting pressure on vessel walls) can induce eNOS and increased mitochondrial biogenesis – another example of hormesis!

(Dikalov SI, Am J Physiol Heart Circ Physiol, 2013, Bailey J, Free Radic Biol Med, 2017; Poburko D. Cell Calcium, 2004)

Mitochondria and heart failure

- Heart failure is associated with a gradual decline in the bioenergetic reserve capacity of the myocardium and decreased mitochondrial biogenesis. ATP levels have been shown to decrease by 30% in heart failure, while in end-stage heart failure, activity rates of Complexes I and IV and several TCA cycle enzymes are markedly decreased.
- Heart failure has been described as described as ‘a systemic mitochondrial cytopathy’.
- Cardiomyocytes undergo metabolic reprogramming involving decreased β-oxidation coupled with intracellular lipid deposition and increased glucose utilisation leading to apoptotic cell death and contractile dysfunction. In addition, the TCA cycle intermediate succinate accumulates in the ischaemic myocardium, inducing oxidative damage at reperfusion.
- The molecular mechanisms underlying metabolic reprogramming in the diseased myocardium include hypoxia-inducible factor 1α (HIF1α), which initiates a transcriptional programme involving PPARγ that leads to increased glucose uptake and consequent lipid accumulation,.
- Diminished redox capacity with lower total glutathione and coenzyme Q10 levels are also a feature of chronic left ventricular failure.

(Rosca MG, Heart Fail Rev, 2013; Bonora M, Nat Rev, 2019)

Other aspects of mitochondria and heart disease

- Animal studies have shown that blocking production of the proteins that stimulate mitochondrial biogenesis led to multiple defects in heart function due to depleted energy reserves. Poor mitochondrial biogenesis led on to heart failure.
- Wolff-Parkinson-White syndrome is now recognised as a mitochondria-associated disorder. A cardiac mitochondrial defect can lead to a sudden conduction velocity disorder and cardiac failure.

(Sheeran FL, Adv Exp Med Biol, 2017; Jiang Y, Toxicology, 2015; Silva-Oropeza E, 2004; Bonora M, Nat Rev, 2019; Bonora M, Nat Rev, 2019; Montaigne D, Card Electrophysiol Clin, 2015; Lee TW, J Biomed Sci. 2017)

Heart failure treatment, courtesy of the Mayo Clinic

- ‘Heart failure is a chronic disease needing lifelong management. However, with treatment, signs and symptoms of heart failure can improve, and the heart sometimes becomes stronger. Treatment may help you live longer and reduce your chance of dying suddenly.
- Doctors sometimes can correct heart failure by treating the underlying cause. For example, repairing a heart valve or controlling a fast heart rhythm may reverse heart failure. But for most people, the treatment of heart failure involves a balance of the right medications and, in some cases, use of devices that help the heart beat and contract properly.’
- The ‘right medication’ (of which patients may have to take several):
  - Angiotensin-converting enzyme (ACE) inhibitors or angiotensin II receptor blockers: vasodilators, decrease workload on the heart.
  - Beta blockers: slows heart rate, reduces blood pressure, limits some of the heart damage.
  - Diuretics or aldosterone antagonists: reduce fluid retention but may alter electrolyte balance.
  - Inotropes or digoxin: improves heart pumping function, strengthening contractions.
- These come with a lot of potential adverse effects.
- Note that nowhere is mentioned that the root cause of heart failure is lack of energy.
- And while we are on the subject of medications…..

Statins: How do they affect the mitochondria?

- **Statins work by blocking HMG-CoA reductase**, the liver enzyme that makes cholesterol; blocking this enzyme triggers **depletion of CoQ10 and vitamin K2**, both necessary for heart health and the prevention of some cancers. The body makes cholesterol as is necessary for the production of hormones etc. https://www.health.harvard.edu/heart-health/how-its-made-cholesterol-production-in-your-body)
- **Statins reduce CoQ10 production, leading to lower ATP production and increased electron leakage from the ETC and hence elevated mitochondrial ROS.**
- A study of rat mitochondria exposed to statins found increased ROS production, **mitochondrial swelling, collapse of mitochondrial membrane potential, cytochrome c release, DNA fragmentation, up to 96% decreased β-oxidation and ATP levels**.
- Some scientists have referred to statins as '**mitochondrial toxins**'.
- Also with statins we also see **incidence of statin-induced myopathy ranging between 5% and 29%.** A 2008 review showed that not only is mitochondrial dysfunction associated with myopathy but also with other adverse effects of statins, such as such as cognitive loss, neuropathy, pancreatic and hepatic dysfunction and sexual dysfunction. The reduced ATP production can lead to worsening of heart disease, particularly heart failure.
- Patients with statin-associated myopathy had increased mitochondrial H2O2 production, Bax/Bcl-2 ratio and caspase 3 cleavage in skeletal muscle, indicating upregulation of the apoptosis pathway.

Statins: do they even help prevent CVD?

- **A 2015 systematic review of statin trials found that in primary prevention trials, the median postponement of death was just 3.2 days. In secondary prevention trials, death was postponed 4.1 days.** This is a truly astounding finding, considering people take statins for years, if not decades, and the fact that these drugs are associated with a wide range of serious side effects.
- Among those with **life-limiting illness, patients who discontinued statins had improved quality of life and fewer cardiovascular events.**
- **Furthermore, a 2015 study showed that statins can actually cause atherosclerosis and heart failure** 'through the depletion of coenzyme Q10…and thereby ATP generation.'
- In 2019, **a group of UK doctors and scientists**, led by Dr Aseem Malhotra and including the editor of the BMJ and the past President of the Royal College of Physicians, **wrote to the Chair of the British Parliamentary Science and Technology Committee,** Sir Norman Lamb MP, who **called for a full investigation into statins.** (https://www.europeanscientist.com/en/features/do-statins-really-work-who-benefits-who-has-the-power-to-cover-up-the-side-effects/)

(Kristensen ML, BMJ Open, 2015; Bouitbir J, Antiox Redox Signal, 2016; Bouitbir J, Eur Heart J, 2012; Tolosa L, Arch Toxicol, 2015; Sirvent P, Toxicol Appl Pharmacol, 2012; Ramachandran R, J Clin Med, 2017; Rundek T, Arch Neurol, 2004 Kaufmann P, Cell Mol Life Sci, 2007; Golomb BA, Am J Cardiovasc Drugs, 2008; Mortensen SA, Mol Aspects Med, 1997; Sadighara M, Basic Clin Pharmacol Toxicol, 2017; Okuyama H, Expert Rev Clin Pharmacol. 2015)

Uffe Ravnskov Review

- A 2018 review of recent clinical trials presents substantial evidence that total cholesterol and LDL cholesterol levels are not an indication of heart disease risk, and that statin treatment is of “doubtful benefit” as a form of primary prevention for this reason.
- The review concludes that statins are unable to satisfy any of the Bradford Hill criteria for causality for heart disease and that the conclusions of authors who find benefit are based on misleading statistics, exclusion of unsuccessful trials and by ignoring numerous contradictory observations.
- If you look at absolute risk, statin drugs benefit just 1% of the treated participants. Out of 100 people treated with statins for five years, one person will have one less heart attack.

(Ravnskov U, Exp Rev Clin Pharmacol, 2018)

Statins double rates of diabetes, a key risk factor for CVD

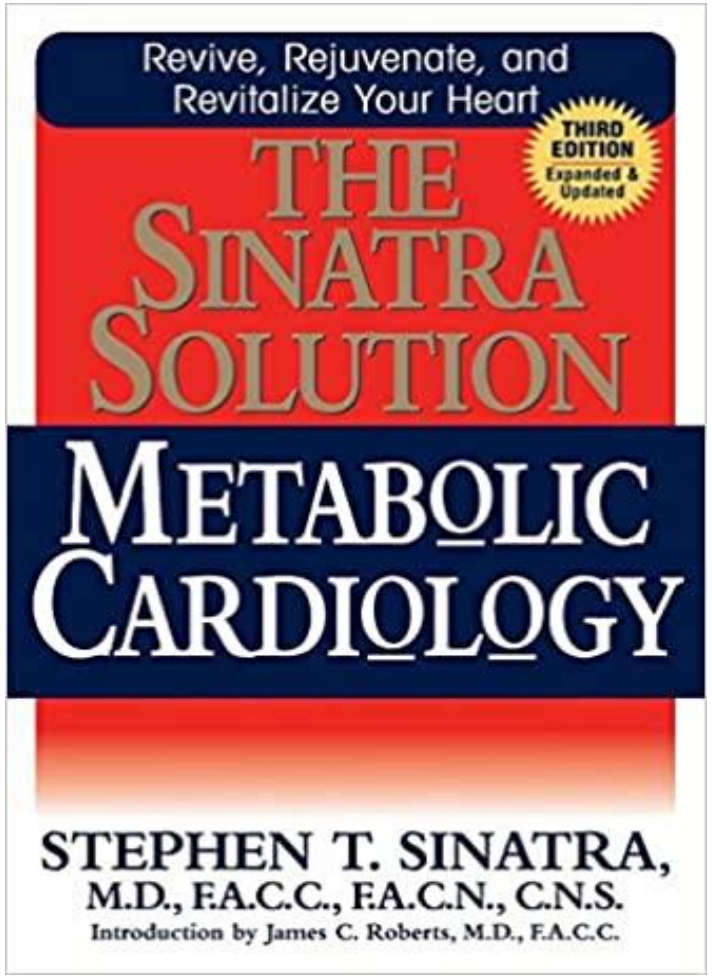
- As long ago as 2012, the US FDA warned of the increased risk of diabetes and impaired glycaemic control in patients who already have diabetes.
- **A 2019 study found that statin users had a 220% higher risk of developing new onset diabetes. Those taking statins for >2 years had a 333% higher risk.** No differences were observed by statin class or intensity of dose. Other studies have broadly confirmed these findings.
- Statins are known to exert side effects on the pancreas, can induce a reduction in insulin secretion and are able to induce apoptosis signalling. Despite this, use of statin therapy in patients with type T2D has been recommended by most clinical guidelines.
- Diabetes UK attempts but fails to reconcile the need to take statins to prevent CAD and diabetic cardiac complications, while potentially causing diabetes in the first place! (https://www.diabetes.co.uk/in-depth/the-convoluted-nature-of-statin-treatment-and-its-link-with-diabetes-2/)

(Elnaem MH, J Pharm Bioallied Sci, 2017; Aiman U, J Pharmacol Pharmacother, 2014; Zigmont VA, Diabetes Metab Res Rev, 2019; Cederberg H, Diabetologia, 2015; Okuyama H, Exp Rev Clin Pharmacol, 2015; Kutner JS, JAMA Intern Med. 2015; Crandall JP, Cardiovascular and Metabolic Risk, 2017; Kim J. Cardiovasc Diabetol, 2018; Shah RV, Circulation, 2012)

Cardiac amyloidosis

- Wikipedia: Cardiac amyloidosis is the deposition of amyloid in the cardiac muscle and surrounding tissues (hear atria, ventricles or valves). Deposits of amyloid, a misfolded and insoluble protein, can cause thickening of different sections of the heart, leading to decreased cardiac function.
- Could cardiac amyloid also be antimicrobial?
- Microbial infection in the heart is known as infective endocarditis, an inflammation of the endocardium.
- What does the literature say?
- 'We report a child whose endocarditis went unrecognised, and who developed amyloidosis' (Herbert MA, Pediatr Nephrol. 1995)
- 'A woman with a diagnosis of bacterial endocarditis of the native aortic valve….staining for amyloid confirmed…amyloidosis with extensive amyloid depositions of the aortic valve and valvular damage.' (van Bentum R, BMC Infect Dis. 2019)
- 'Streptococcus mutans can cause infectious endocarditis….. Streptococcus mutans biofilms contain amyloid fibrils.' (Paranjapye N, J Mol Biol. 2018)

Dr Stephen Sinatra



Dr Stephen Sinatra: early contribution to understanding the importance of mitochondria in the heart

- Stephen Sinatra was one of the first cardiologists to appreciate the crucial importance of mitochondrial energy production in the heart to keep blood pulsing through the body.
- **He points out that levels of adenine nucleotides (ATP, ADP, AMP) decline more than 30% in heart failure and more than 40% in coronary artery disease and ischaemic heart disease.**
- Stephen Sinatra's principal focus is on **heart failure**, for which **conventional medicine has no cure, only symptomatic relief**.
- He regularly prescribes the '**awesome foursome**' (coenzyme Q10, L-carnitine, magnesium and D-ribose) **for almost any form of CVD**.
- His 2009 review (Sinatra ST, Alternative Therapies, 2009) talks of how understanding Bioenergetics (the study of energy transformation in living organisms) is critical for optimising both ATP concentrations in the cell and the efficiency of ATP turnover and recycling.

Dr Stephen Sinatra on heart failure

- Heart failure simply means that the heart is unable to pump blood around the body properly due to energy starvation. Heart failure is typically associated with an overstretched, thickened and enlarged left ventricle or left ventricular hypertrophy (LVH), a situation that overtaxes the heart muscle with each contraction.
- Heart failure is associated with a gradual decline in the bioenergetic reserve capacity of the myocardium and decreased mitochondrial biogenesis. In end-stage heart failure, activity rates of Complexes I and IV and several TCA cycle enzymes are markedly decreased.
- In heart failure, levels of adenine nucleotides (ATP, ADP, AMP) decline more than 30% (and more than 40% in coronary artery disease and ischaemic heart disease). Tissue biopsy and nuclear magnetic resonance findings confirm these observations.
- Sinatra says that in part, the disparity between energy supply and demand is due to a shift in the relative contributions of fatty acid vs glucose oxidation to ATP synthesis. Where the main ATP production is via carbohydrate metabolism, the total capacity for ATP synthesis decreases, but all the time demand is increasing.

(Sinatra ST, Alternative Therapies, 2009)

Dr Stephen Sinatra: Coenzyme Q10

- **Sinatra has published 4 papers showing a correlation between levels of CoQ10 and severity of heart failure and the value of supplementing CoQ10.**
- **After the latest paper (2004) he gave up as he realised nothing was going to change medical opinion. Apparently less than 1% of good medical research makes it into clinical practice.**
- **Many studies have shown deficiencies of CoQ10 in all forms of CVD** (heart failure, hypertension, aortic valve disease, mitral valve prolapse, cardiomyopathy and coronary artery disease and following bypass surgery). Furthermore, there is plenty of scientific opinion in favour of using CoQ10 in heart failure and dilated cardiomyopathy as it improves left ventricular function, diastolic dysfunction, ejection fraction, exercise tolerance, clinical outcome and quality of life.
- **Furthermore its production is inhibited in anyone on statins** i.e. many people over the age of 50 and all of the above! Statins inhibit HMG-CoA reductase, the enzyme that makes both cholesterol and CoQ10.
- Sinatra's rationale for using CoQ10 for heart patients is principally because it is a key component of Complex II of the ETC and increases ATP production, which has a positive impact on diastolic function.
- Other benefits: it is an excellent antioxidant for lipids, particularly preventing oxidation of LDL cholesterol, reduces platelet size and limits their aggregation, stabilises cell membranes (reducing risk of arrhythmia).

(Sinatra ST, Clin Cardiol, 2004; Sinatra ST, Ann Intern Med, 2000; Sinatra ST, Conn Med, 1997; Sinatra ST, Mol Aspects Med, 1997; Sinatra ST, Alternative Therapies, 2009)

Dr Stephen Sinatra: Coenzyme Q10 supplementation

- In Sinatra's opinion, **supplementing CoQ10 should be the first defence against heart failure or dilated cardiomyopathy.**
- Bioavailability can be a problem; be sure to use a quality lipid-based form or use **ubiquinol**, the reduced form, if there is severe energy depletion.
- **For dosage Sinatra recommends being guided by blood levels:** For severely diseased hearts, 3.5µg/ml. In other patients, 2.5 µg/ml may be sufficient.
- In an analysis in 3 patients with refractory heart failure, these higher doses of CoQ10 were required in order to get a therapeutic result.
- Dosage:
  - prevention: 90-150 mg/day
  - angina, arrhythmia, hypertension and patients on statins: 180-360 mg/day
  - mild/moderate heart failure or ischaemia: 300-360 mg/day
  - severe heart failure, dilated cardiomyopathy: 360-600 mg/day

Dr Stephen Sinatra: L-carnitine

- **Fatty acids are the heart's preferred energy source, but this requires an adequate supply of L-carnitine.** L-carnitine is the rate-limiting factor in utilisation of fatty acids.
- **L-carnitine not only promotes OXPHOS by providing fats for β-oxidation, it also helps clear lactic acid**, acts as an antioxidant, promotes vasodilation, facilitates the production and utilisation of ketones and protects against ammonia toxicity. L-carnitine also plays a significant role in the detoxification pathways in the mitochondria, enhancing the turnover of ATP, thus supporting diastolic function.
- **Annex F** shows lots of studies of L-carnitine: A meta-analysis shows that L-carnitine improves overall efficacy, left ventricular ejection fraction, strike volume and cardiac output in heart failure, another shows that L-carnitine induces a significant reduction in all-cause mortality, ventricular arrhythmias and development of angina in acute myocardial infarction and a third meta-analysis shows that in patients with dilated cardiomyopathy, L-carnitine was associated with improvement in overall efficacy, left ventricular ejection fraction and cardiac output, with significantly decreased left ventricular end-diastolic dimension (Song X, Biomed Res Int. 2017; DiNicolantonio JJ, Mayo Clin Proc. 2013; Weng Y, Biomed Res Int. 2021)
- **Patients with heart failure and dilated cardiomyopathy were given 2g/day L-carnitine vs placebo; after a mean 34 months, 18.1% of the placebo group had died compared to 2.7% of the L-carnitine group**. Kaplan-Meier survival analysis showed that patients' survival on L-carnitine was statistically significant. (Rizos I. Am Heart J. 2000)

Dr Stephen Sinatra: L-carnitine supplementation

- **L-carnitine deficiency is commonly seen in heart disease patients** and also with ageing, co-factor deficiencies (vitamin C, B vitamins, iron) and in vegetarians/vegans. An L-carnitine deficiency sets up a vicious cycle of deficiency.
- **Sinatra uses L-carnitine to help patients with angina, heart failure, peripheral vascular disease and arrhythmia.**
- **L-carnitine also lowers total and LDL cholesterol and triglycerides and raises HDL cholesterol levels by activating lipoprotein lipase.** It works in synergy in the heart with CoQ10.
- Dosage:
  - Prevention: 250-750 mg/day
  - Hypertension, mitral valve prolapse 500-1000 mg/day
  - Stable angina, arrhythmia, atrial fibrillation, heart failure 1000-2000 mg/day
  - Severe heart failure 2500-3500 mg/day in divided doses

Dr Stephen Sinatra: Magnesium

- In the cell, **magnesium is concentrated in the mitochondria and attaches to ATP to activate it**.
- **Magnesium is a natural calcium channel blocker,** can be anti-arrhythmic, improve the LDL/HDL ratio, act as a vasodilator, inhibit clot formation, MI and stroke, relieve symptoms of mitral valve prolapse, reduce heart failure, prevent atherosclerosis and maintain vascular tone.
- **Take at least 400mg/day as a preventive, higher doses if there is pre-existing CVD.** There is no toxicity with a high magnesium intake.
- As with calcium, magnesium stores are contained in the bones. So any prolonged magnesium deficiency is going to result in weaker bones. Anyone or taking proton pump inhibitors (to reduce acid reflux) and some other medications will have impaired magnesium absorption.

Dr Stephen Sinatra – D-Ribose

But first we must talk about ATP:ADP:AMP balance

Healthy hearts:

- ATP, ADP and AMP (all adenine nucleotides) are maintained in a **fairly constant ratio**: normally there is roughly 10 times more ATP than ADP and about 100 times more ATP than AMP. ADP and Pi will be recycled immediately back to ATP to continue energy production.

Oxygen-starved or diseased hearts:

- **With lack of oxygen for OXPHOS, the cell reverts to anaerobic glycolysis, with glucose/pyruvate being metabolised to make ATP and lactic acid through,** causing an increase in cellular acidity. This **acidity signals that more ATP is required,** but a failing heart will be unable to deliver.
- When **AMP, ADP and ATP are not being utilised, they leak out of the cell, so the means of producing more ATP is lost.**
- Just 2 hours of transient hypoxia can deplete nucleotide reserves by 15% but **in heart failure, ischaemic heart disease and cardiomyopathy, the depletion can be 50%.**

How to rebuild adenine nucleotides: AMP

- **AMP comprises adenine, ribose and 1 phosphate group**. There is not normally a shortage of phosphate groups, while **adenine (a purine derivative) and ribose may be in short supply.**
- The purine pool is a collection of molecules that drives the combination of 2xADP to form 1xATP and 1xAMP. **Rebuilding the stock of purines can be a slow process and requires the 5-carbon sugar D-ribose. The availability of D-ribose is the rate-limiting factor in rebuilding AMP.**
- **Ribose** is synthesised through 2 possible pathways:
  - **Purine salvage** – the degradation of AMP leaves behind adenine, inosine and hypoxanthine, which, given enough time, can combine to form D-ribose. **The slow pathway**; if a heart suffers ischaemia, it can take nearly **10 days** to rebuild the purine pool.
  - ***De novo* purine synthesis** – **creating D-ribose from glucose from scratch via the pentose phosphate pathway.** But if the heart is starved of oxygen and energy production transfers from OXPHOS to glycolysis, then all available glucose will be utilised for this and will not be available for the pentose phosphate pathway. **The very slow pathway:** it would take the human heart over **100 days** to make all its energy molecules this way. And in sick patients, this pathway will be even slower.
- Purines (and pyrimidines) make up the two groups of nitrogenous bases, adenine, guanine and hypoxanthine, which also contribute to DNA and RNA and other high-energy molecules. They are normally found with a pentose (5-carbon) sugar (such as ribose) attached to the nitrogen atom to form a nucleoside.
- The diet provides negligible amounts of purines as they are broken down in the gut to form uric acid. So eating more is not going to help – may cause gout!

Ribose supplementation to the rescue!

- **Supplementing D-ribose can rebuild the purine pool within 2 days**. It is the only molecule that can rapidly restore the purine pool and hence energy reserves. The heart's ability to resynthesise ATP and restore the depleted energy pool in limited by the availability of D-ribose.
- **No drugs have yet been found to increase the cardiac purine pool.**
- **Sinatra recommends D-ribose for any form of angina, heart failure and dilated cardiomyopathy and in any patients on inotropic drugs** as they place a great strain on the heart by forcing it to beat more strongly and cause depletion of energy reserves. Supplementing ribose will provide additional ATP and avoid the painful lactate production.
- **Take up to 15g in 3 divided doses with meals.**
- D-ribose administration bypasses the rate-limiting steps of the pentose phosphate pathway, accelerating myocardial adenine nucleotide synthesis, thereby increasing contractile reserve to aid recovery of cardiac diastolic performance.
- [Note: Other sites (but not Dr Sinatra) say that patients taking warfarin should not take ribose.]

Dr Stephen Sinatra: Other recommended remedies

- Hypertension: omega-3 fish oil 3g/day; nattokinase 100mg/day; garlic 1g/day; hawthorne 1000-1500mg/day
- Stable angina: vitamin K2 150mcg/day (as MK7)
- Arrhythmia: intermittent atrial fibrillation: omega-3 fish oil 4g/day
- Heart failure: omega-3 fish oil 1g/day

Cardiovascular disease and mitochondrial therapies

- **Caloric restriction (CR)/fasting**: Many human studies showing efficacy for heart failure, diastolic dysfunction, hypertension and increasing LDL particle size.
- **Ketogenic diet**: Significant decrease in mortality, stroke (for saturated fat), blood pressure, total/LDL cholesterol and triglycerides and increased HDL cholesterol. A 2017 18-country **Lancet study called for revision of dietary guidelines** (Dehghan M, Lancet, 2017)
- **Exercise**: Both aerobic and resistance exercise improved blood pressure, while aerobic exercise also improved lipid profile, heart rate, carotid-femoral pulse wave velocity, cardiac output and left ventricular ejection fraction.
- Hyperbaric oxygen: mixed results.
- Hypothermia: mixed results.
- **Near infrared radiation**: few human studies but a systematic review of animal studies shows benefit in infarct size (up to 76% reduction), decreased inflammation and scarring, and increased tissue repair.
- Pulsed electromagnetic fields: few human studies.

Fasting or caloric restriction (CR) and CVD

- A 2020 systematic review showed that CR improved efficiency of myocardial tissue in heart failure patients with and without preserved ejection fraction (Bianchi DE, Clin Nutr ESPEN, 2020).
- In subjects with and without T2D, CR improved cardiac function, decreased myocardial fatty acid uptake, myocardial mass and cardiac work; it also improved heart rate variability, isovolumic relaxation time, ventricular stiffness, septal annulus motion and left ventricular diastolic function (van Eyk HJ, Nutr Diabetes, 2018; Viljanen AP, Am J Cardiol, 2009; Ito H, Jpn Heart J, 2001; Riordan MM, Am J Physiol Heart Circ Physiol, 2008; Meyer TE, JACC, 2008)
- A 2018 review found that CR improved hypertension, fast heart rate, low heart rate variability, sympathetic nervous system dominance over parasympathetic, arterial stiffness, endothelial dysfunction and poor flow-mediated arterial dilatation, all associated with cardiovascular mortality and morbidity. It is effective regardless of age, gender, ethnicity, weight, BMI, metabolic syndrome or T2D. (Nicoll R, Int J Mol Sci, 2018)
- CR or intermittent or alternate day fasting (ADF) lowered systolic blood pressure (SBP) and increased low density lipoprotein (LDL) particle size, rendering it less atherogenic, and enhanced arterial compliance but its effect on blood lipids varied (Hoddy KK, Obesity, 2014; Zuo L, Front Physiol, 2016; Weiss EP, Am J Clin Nutr, 2016; Bhutani S, Obesity, 2010).

Ketogenic diet and CVD

- Overweight or obese subjects placed on a ketogenic diet showed a significant decrease in total and LDL cholesterol and triglycerides and an increased in HDL cholesterol (Paoli A, Mar Drugs, 2015; Dashti HM, Mol Cell Biochem, 2006).
- A ‘Mediterranean’ ketogenic diet resulted in decreased total and LDL cholesterol and triglycerides, decreased systolic and diastolic blood pressure and a significant increase in HDL cholesterol in overweight or obese subjects (Paoli A, Nutr J, 2011; Pérez-Guisado J, Nutr J, 2008).
- An 18-country study showed that high carbohydrate intake was associated with higher risk of total mortality, whereas fat intake was associated with lower total mortality. Saturated fat showed an inverse association with stroke. No association of total fat and fat types with CVD, MI, and CVD mortality. (Dehghan M, Lancet, 2017)

Saturated fat and CVD

- The recommendation to limit dietary saturated fatty acid (SFA) intake has persisted despite mounting evidence to the contrary. Most recent meta-analyses of randomized trials and observational studies found no beneficial effects of reducing SFA intake on cardiovascular disease (CVD) and total mortality, and instead found protective effects against stroke. Although SFAs increase low-density lipoprotein (LDL) cholesterol, in most individuals, this is not due to increasing levels of small, dense LDL particles, but rather larger LDL particles, which are much less strongly related to CVD risk. It is also apparent that the health effects of foods cannot be predicted by their content in any nutrient group without considering the overall macronutrient distribution. Whole-fat dairy, unprocessed meat, and dark chocolate are SFA-rich foods with a complex matrix that are not associated with increased risk of CVD. The totality of available evidence does not support further limiting the intake of such foods. (Astrup A, J Am Coll Cardiol, 2020)
- Meanwhile the American Heart Association website recommends 'Use cooking oils that are lower in saturated fat. Good choices include avocado, canola, corn, grapeseed, olive, peanut, safflower, sesame, soybean and sunflower oils.'
- The UK government Scientific Advisory Committee on Nutrition (SACN) recently published a review of dietary fats and health and recommended that saturated fats should not exceed >10% of total energy. !0% is a completely arbitrary figure and has no basis in science.
- Meanwhile the British Heart Foundation has a downloadable leaflet entitled 'Taking control of saturated fats'. It start off with 'Reducing the amount of saturated fat you eat is a very simple way to lower your cholesterol level and support your heart health'.

Exercise and CVD

- A 2018 meta-analysis found that both aerobic and resistance exercise over 24 weeks in subjects aged between 50-60 reduced aortic systolic pressure, while resistance exercise decreased aortic diastolic pressure and aerobic exercise decreased the augmentation index and improved carotid-femoral pulse wave velocity, cardiac output and left ventricular ejection fraction (Zhang Y, PLoS One, 2018).
- Two further 2018 meta-analyses found that aerobic exercise improved HDL, triglycerides, diastolic blood pressure and cardiorespiratory fitness, particularly with vigorous intensity and conducted 3 days/week for ≥12 weeks; no significant effects were determined following resistance exercise. In untrained women, aerobic exercise exercise lowered heart rate, systolic blood pressure and total and LDL cholesterol. (Wewege MA, Atherosclerosis, 2018; Zhang Y, J Sports Med Phys Fitness, 2018)
- Meta-analyses of heart failure patients showed that exercise improved maximum heart rate, diastolic function, walking speed, peak oxygen consumption and quality of life (Dieberg G, J Appl Physiol, 2015; Ismail H, JACC Heart Fail, 2013).
- Two 2019 meta-analyses found that in patients with intermittent claudication, supervised exercise lowered systolic and diastolic blood pressure and total LDL cholesterol, with no effect on heart rate, triglycerides or HDL (Cornelis N, Eur J Vasc Endovasc Surg, 2019; Jansen SCP, J Vasc Surg, 2019)
- A 2017 meta-analysis of patients with stoke or TIA, exercise reduced systolic blood pressure and increased HDL cholesterol (D'Isabella NT, Clin Rehabil, 2017).

Photobiomodulation

- A growing body of evidence supporting the use of photobiomodulation in myocardial infarct models has implicated multiple molecular interactions.
- A systematic review showed that photobiomodulation in animal models showed consistently positive effects over a range of wavelengths and application parameters, with reductions in total infarct size (up to 76%), decreases in inflammation and scarring, and increases in tissue repair.
- Multiple molecular pathways were identified, including modulation of inflammatory cytokines, signalling molecules, transcription factors, enzymes and antioxidants.

(Liebert A, Sci Rep, 2017; Carlos FP, 2016, Life Sci)

Mitochondrial remedies for which there is human evidence (Annex F)

- B vitamins
- L-carnitine
- Coenzyme Q10
- Lipoic acid
- Magnesium
- Melatonin
- Omega 3 fatty acids
- D-Ribose
- Taurine
- Vitamin D
- Curcumin
- Grape seed extract
- Quercetin
- Resveratrol
- Genistein