INTEGRIA HEALTHCARE SYMPOSIUM 2015

Clinical Pearls + Practitioner Perspectives

An Experience-Based Approach to Patient Care

24+25 OCTOBER
MELBOURNE

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DAY 2

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Clinical Pearls + Practitioner Perspectives

An Experience-Based Approach to Patient Care

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Clinical Pearl:
('klɪnɪkəl pûrl)

1. A straightforward and meaningful piece of clinical advice
2. Clinically relevant information based on experience

Session 5

The Cardiometabolic Continuum: The Nutritional Medicine Perspective

DAY 2

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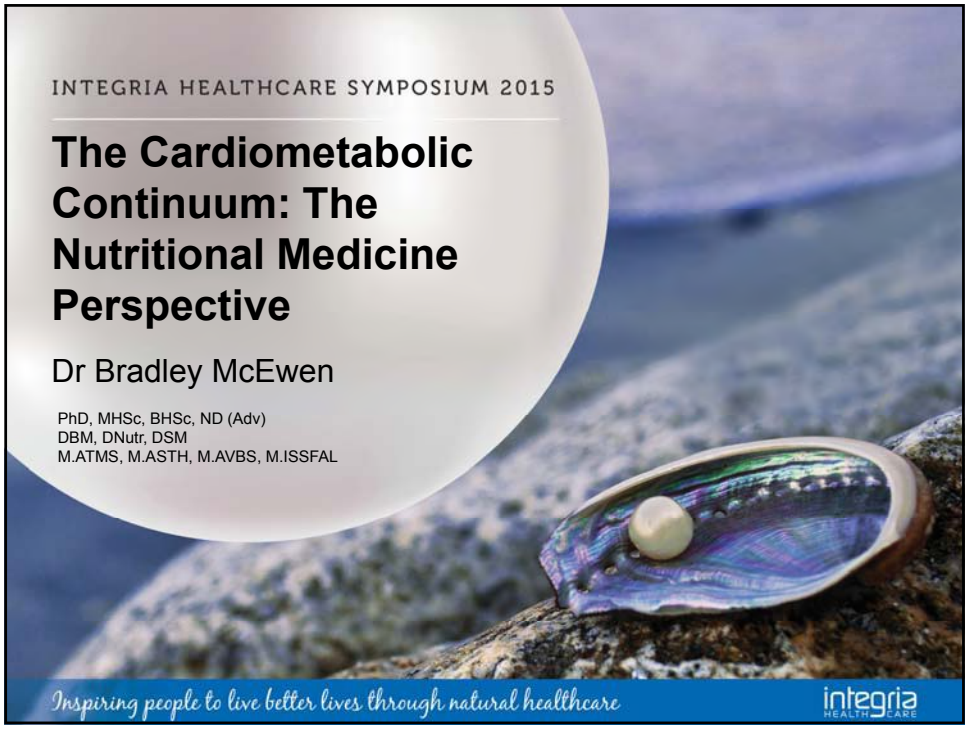


Key Abbreviations

- ABS – Australian Bureau of Statistics
- ADP – adenosine diphosphate
- BMI – body mass index
- BP – blood pressure
- CAD – coronary artery disease
- CHD – coronary heart disease
- COX-2 – cyclooxygenase-2
- CRP – C-reactive protein
- CVD – cardiovascular disease
- DHF – dihydrofolate
- ECM – extracellular matrix
- eNOS – endothelial nitric oxide synthase
- GI – glycaemic index
- GL – glycaemic load
- GLUT – glucose transporter
- GP – glycoprotein
- HbA1c – glycated haemoglobin
- HDL – high-density lipoprotein/s
- Hg – mercury
- HMG-CoA – 3-hydroxy-3-methylglutaryl coenzyme A
- HOMA-IR – homeostasis model assessment-estimated insulin resistance
- IL – interleukin

integria HEALTH CARE 2

Key Abbreviations

- LDL – low-density lipoprotein/s
- MI – myocardial infarction
- MMP-9 – matrix metallopeptidase 9
- MTHFR – methyltetrahydrofolate reductase
- NAD – nicotinamide adenine dinucleotide
- NFkB – nuclear factor kappa beta
- Nrf2 – nuclear factor erythroid 2–related factor 2
- PAI-1 – plasminogen activator inhibitor-1
- PCOS – polycystic ovary syndrome
- PPARγ – peroxisome proliferator-activated receptor gamma
- PUFA – polyunsaturated fatty acids
- ROS – reactive oxygen species
- SAH – S-adenosylhomocysteine
- SAMe – S-adenosyl-methionine
- SMC – smooth muscle cells
- THF – tetrahydrofolate
- TNF-α – tumour necrosis factor-α
- TXA – thromboxane
- VCAM-1 – vascular cell adhesion molecule 1
- WHR – Waist-hip ratio

Leading Causes of Death: Australia 2013

- In 2013, the leading underlying cause of death for all Australians was *ischaemic heart disease*, which includes angina, blocked arteries of the heart and heart attacks
- Ischaemic heart diseases were identified as the underlying cause of 19,766 deaths, 13.4% of all deaths registered in 2013
- The proportion of deaths due to ischaemic heart diseases has decreased over the past 10 years, from 18.5% (24,576) in 2004 to 13.4% (19,766) in 2013
- The top 10 leading causes of death accounted for 51.2% of all deaths registered in 2013, and the top 20 leading causes accounted for 67.0% (ABS 2015)

ABS (Australian Bureau of Statistics), 3303.0 - Causes of Death, Australia, 2013, http://www.abs.gov.au/ausstats/abs@.nsf/mf/3303.0, updated 31/3/2015, viewed 06/07/2015

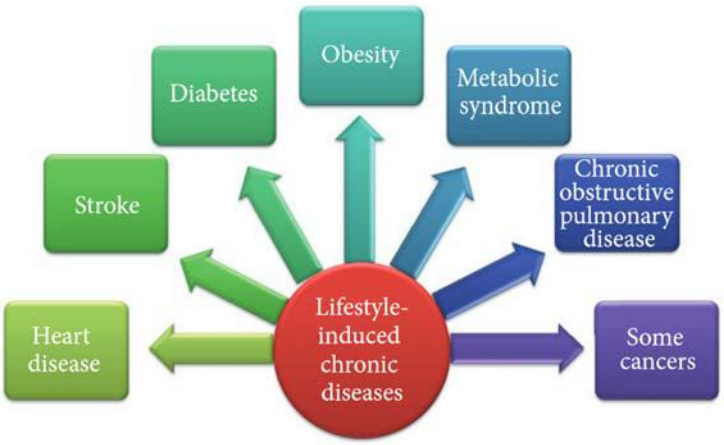
Leading Causes of Death, Australia – Selected Years: 2004, 2008, 2013

	2004		2008		2013		
Cause of death	**Number**	**Rank**	**Number**	**Rank**	**Number**	**Rank**	↑↓↔
Ischaemic heart diseases	24,576	1	23,813	1	*19,766*	*1*	↔
Dementia and Alzheimer disease	4,606	5	8,172	3	*10,993*	*2*	↑
Cerebrovascular diseases	12,041	2	11,979	2	*10,549*	*3*	↓
Trachea, bronchus and lung cancer	7,264	3	7,956	4	8,217	4	↔
Chronic lower respiratory diseases	5,785	4	6,255	5	7,148	5	↔
Diabetes	3,599	8	4,181	6	*4,328*	*6*	↑
Colon, sigmoid, rectum and anus cancer	4,126	6	4,125	7	4,234	7	↔
Blood and lymph cancer (including leukaemia)	3,820	7	3,887	8	4,094	8	↔
Heart failure	2,823	11	3,363	9	*3,244*	*9*	↑
Prostate cancer	2,761	12	3,031	11	3,112	10	↑

Leading Causes of Death, Australia – Selected Years: 2004, 2008, 2013

	2004		2008		2013		
Cause of death	**Number**	**Rank**	**Number**	**Rank**	**Number**	**Rank**	↑↓↔
Diseases of the urinary system	2,896	10	3,235	10	2,987	11	↓
Breast cancer	2,661	13	2,789	12	2,892	12	↔
Pancreatic cancer	1,978	15	2,289	14	2,558	13	↓
Intentional self-harm	2,098	14	2,340	15	2,520	14	↑
Influenza and pneumonia	3,381	9	1,760	17	2,493	15	↑
Skin cancers	1,573	16	1,857	15	2,209	16	↓
Hypertensive diseases	1,340	18	1,833	16	*2,150*	*17*	↓
Accidental falls	873	20	1,461	20	1,920	18	↑
Cardiac arrhythmias	1,229	19	1,550	18	*1,892*	*19*	↓
Cirrhosis and other diseases of liver	1,386	17	1,509	19	*1,772*	*20*	↓
Total deaths	**90,816**		**97,385**		**99,078**		↑

Lifestyle-Induced Chronic Disease



Minich DM, Bland JS. *Scientific World Journal*. 2013; 2013: 129841

The Cardiometabolic Syndrome

- The cardiometabolic syndrome is a constellation of maladaptive abnormalities:
  - cardiovascular
  - metabolic
  - prothrombotic
  - inflammatory
  - renal
- The cardiovascular and metabolic derangements individually and interdependently lead to a substantial increase in cardiovascular disease (CVD) morbidity and mortality
- The cardiometabolic syndrome is an established and strong risk factor for premature and severe cardiovascular disease and stroke

Castro JP, El-Atat FA, McFarlane SI et al. *Curr Hypertens Rep*. 2003 Oct; **5**(5): 393-401

Metabolic Syndrome: Introduction

- Metabolic syndrome is a multicomponent risk factor for cardiovascular disease and type 2 diabetes that reflects the clustering of individual cardiometabolic risk factors related to abdominal obesity and insulin resistance

Mozaffarian D, Benjamin EJ, Go AS et al. *Circulation*. 2015; **131**(4): e29-e322

Metabolic Syndrome: Criteria

- Metabolic syndrome is diagnosed when any 3 of the following 5 risk factors are present:
  - Fasting plasma glucose ≥5.5 mmol/L or undergoing drug treatment for elevated glucose
  - HDL cholesterol <1.0 mmol/L in men or <1.3 mmol/L in women or undergoing drug treatment for reduced HDL cholesterol
  - Triglycerides ≥1.7 mmol/L or undergoing drug treatment for elevated triglycerides
  - Waist circumference >102 cm in men or >88 cm in women for Caucasian people
  - BP ≥130 mm Hg systolic or ≥85 mm Hg diastolic or undergoing drug treatment for hypertension or antihypertensive drug treatment in a patient with a history of hypertension

Mozaffarian D, Benjamin EJ, Go AS et al. *Circulation*. 2015; **131**(4) :e29-e322

Metabolic Syndrome and Other Conditions

- There are many adverse health conditions that are related to metabolic syndrome but are not part of its clinical definition. These conditions include:
  - Non-alcoholic fatty liver disease
  - Polycystic ovarian syndrome (PCOS)
  - General pro-inflammatory state
  - Prothrombotic state
  - Sexual dysfunction (erectile dysfunction in men)
  - Obstructive sleep apnoea
  - Certain forms of cancer
  - Possibly osteoarthritis

Yen CH, Yang NC, Lee BJ et al. *ScientificWorldJournal.* 2013; **3**;2013: 767968

Metabolic Syndrome

- The *Australian Diabetes, Obesity and Lifestyle study (AusDiab)* in 2000, found that 19% of Australians aged 25 years and over met the criteria for a diagnosis of the metabolic syndrome
- The follow up study in 2004–05 found the annual incidence of metabolic syndrome to be 3%
- About a fifth of the world's adult population are thought to have metabolic syndrome

Buitrago-Lopez A et al. *BMJ.* 2011; **343**: d4488
Harris MF. *Aust Fam Physician.* 2013; **42**: 524-527

Cardiovascular Disease: Risk Factors

- Risk factors for CVD include:
  - Platelet hyperaggregation
  - Increased coagulation factor activity
  - Decreased fibrinolytic capacity
  - Diabetes
  - Dyslipidaemia
  - Hypercholesterolaemia (total cholesterol and LDL)
  - Hypertriglyceridaemia
  - Overweight/obesity
  - Nutrition and diet quality
  - Physical inactivity
  - Family history of CVD

References for Previous Slide

Koenig W. *Eur Heart J*. 1998; **19** Suppl C:C39-43
Lorenzet R et al. *Thromb Res*. 2012; **129**(3):285-9
Mozaffarian D et al. *Circulation*. 2015; **131**(4):e29-e322
Ruggeri ZM. *Nat Med*. 2002; **8**(11):1227-34

Metabolic Syndrome and Inflammation

- Metabolic syndrome has been associated with a state of chronic, low-grade inflammation
- There is speculation that low-grade inflammation underlies or exacerbates metabolic syndrome
- Inflammatory cytokines reportedly induce insulin resistance in both adipose tissue and muscle
- In the presence of obesity, adipose tissue produces cytokines in excess, whereas output of adiponectin is reduced
- These responses appear to heighten the connection between obesity and inflammation
- Ectopic lipid accumulation in muscle and liver potentially predisposes to insulin resistance and dyslipidaemia

Grundy SM, Cleeman JI, Daniels SR et al. *Circulation*. 2005; **112**(17): 2735-52

Metabolic Syndrome and Inflammation

- Inflammatory markers are predictors of cardiovascular events and progression to type 2 diabetes in healthy individuals as well as those with the metabolic syndrome
- This highlights the link between inflammation, metabolic disorders, and cardiovascular disease

Sola S, Mir MQ, Cheema FA, Khan-Merchant N et al. *Circulation*. 2005; **111**(3): 343-8

Metabolic Syndrome and Oxidative Stress

- The below markers of metabolic syndrome may increase oxidative stress and reduce antioxidant defences:
  - insulin resistance
  - type 2 diabetes
  - hypertension
  - dyslipidaemia
  - visceral obesity

Yen CH, Yang NC, Lee BJ et al. *ScientificWorldJournal*. 2013; **3**;2013 :767968

Metabolic Syndrome and Oxidative Stress

- Increases in oxidative stress contribute to:
  - impaired vascular function
  - inflammation
  - thrombosis
  - atherosclerosis
- These ultimately give rise to vascular disease

Yen CH, Yang NC, Lee BJ et al. *ScientificWorldJournal*. 2013; **3**;2013: 767968

Oxidative Stress

- Reactive oxygen species (ROS) are a family of highly reactive species that are formed either enzymatically or non-enzymatically causing cell damage either directly or through behaving as intermediates in diverse cell signalling pathways
- Oxidative stress occurs when there is an imbalance between the generation of ROS and the antioxidant defence systems in the body
- The antioxidant system can become overwhelmed

Rodrigo R et al. *Biomed Res Int.* 2013; 2013: 43761.

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Oxidative Stress

- Antioxidant defences can be divided into enzymatic and non-enzymatic.
- Enzymatic antioxidant defences mainly include:
  - Superoxide dismutase
  - Glutathione peroxidase
  - Catalase
  - Thioredoxin peroxidase

Rodrigo R et al. *Biomed Res Int.* 2013; 2013: 437613

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Oxidative Stress

- Non-enzymatic antioxidant defences include:
  - Ascorbic acid
  - (α-) tocopherol
  - Coenzyme Q10
  - Cysteine
  - Flavonoids
  - Carotenoids
  - Reduced glutathione (GSH)
  - Polyphenols

Rodrigo R et al. *Biomed Res Int.* 2013; 2013: 437613

Oxidative Stress

- Polyphenols act as antioxidants both through the prevention of damage from ROS and their iron chelating ability
- The first line of cellular defence against oxidative injury in the heart and most tissues includes the antioxidant enzymes:
  - Catalase
  - Superoxide dismutase
  - Glutathione peroxidase

Rodrigo R et al. *Biomed Res Int.* 2013; 2013: 437613

Oxidative Stress and Inflammation

- Studies suggest that pro-oxidative and pro-inflammatory processes play a significant role in the development of:
  - endothelial dysfunction
  - progression of atherosclerosis
- Chronic inflammation and an abnormal pro-oxidant state are both found in metabolic syndrome and may play a role in its pathogenesis

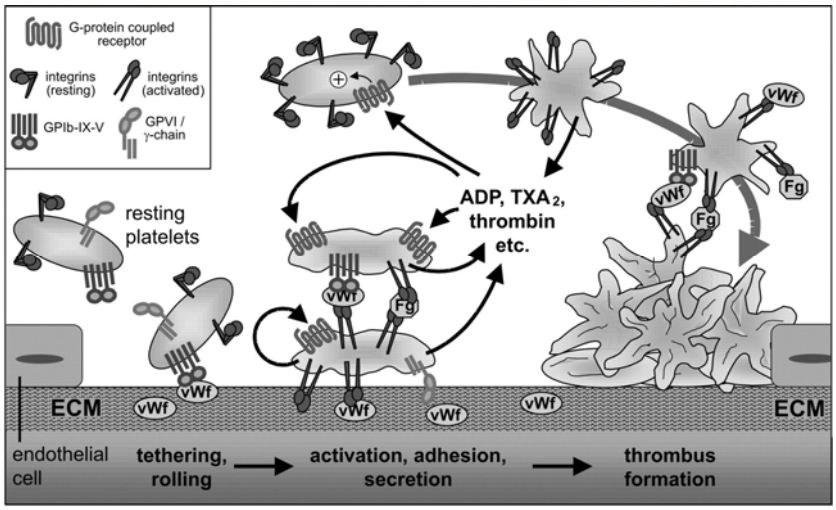
Sola S, Mir MQ, Cheema FA, Khan-Merchant N et al. *Circulation*. 2005; **111**(3): 343-8

Metabolic Syndrome and Platelet Function

- Platelets play a key role in preventing blood loss after injury.
- Platelets are also central in the formation of pathological thrombi responsible for the acute clinical manifestations of atherothrombotic disease

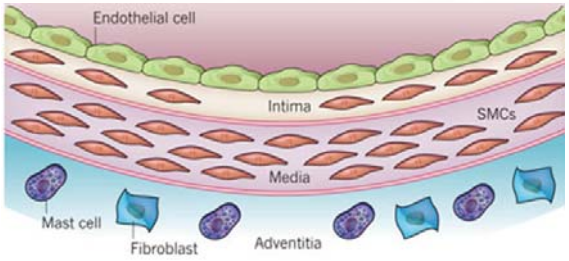
Angiolillo DJ et al. *Eur Heart J*. 2010; **31**(1):17-28

Activation of Platelets at Sites of Vascular Injury



Offermanns S. *Circ Res*. 2006; **99**(12): 1293-304

Stages in the Development of Atherosclerotic Lesions

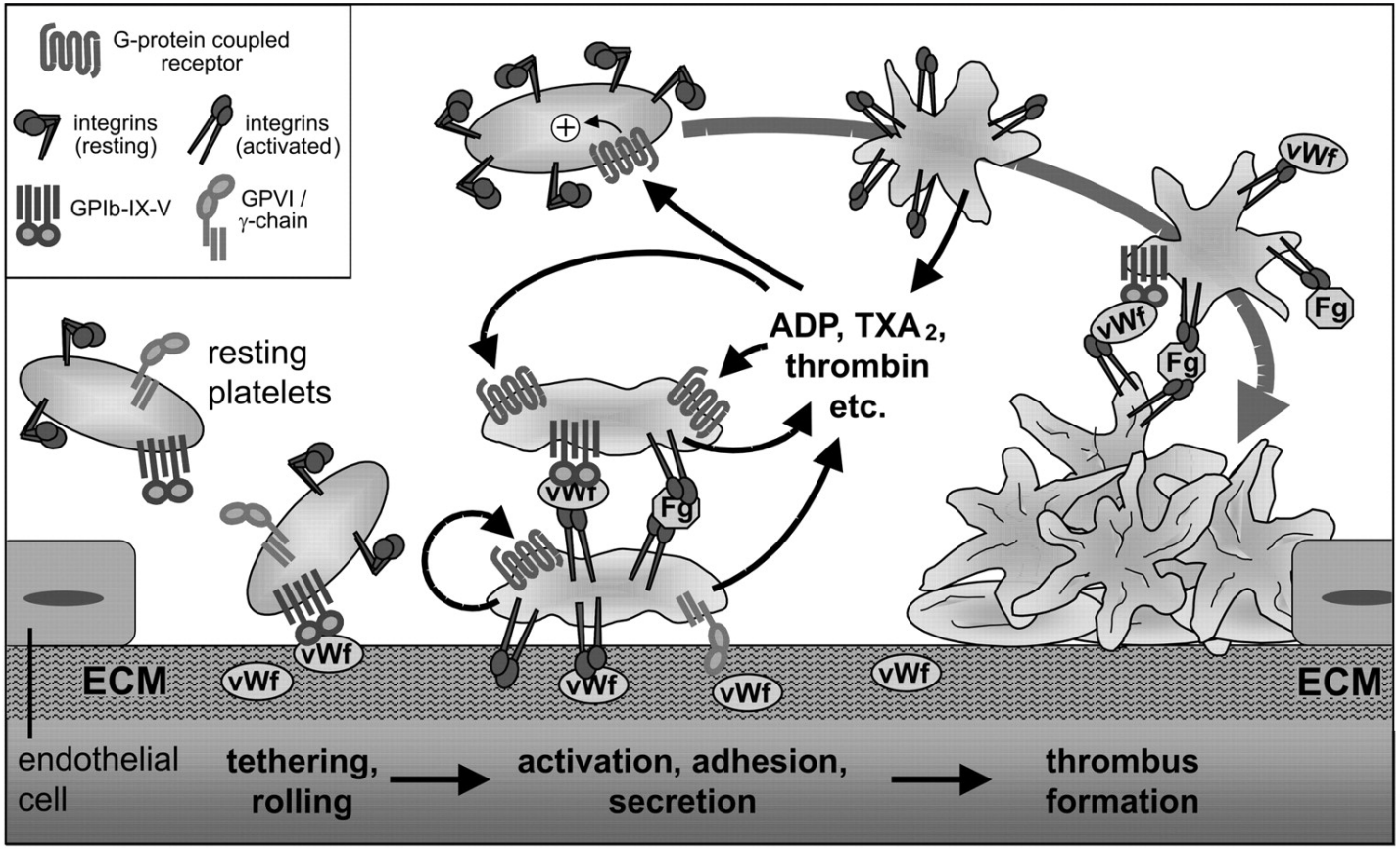


The normal artery contains three layers:

- The inner layer, the tunica intima, is lined by a monolayer of endothelial cells that is in contact with blood overlying a basement membrane
- The human intima contains resident smooth muscle cells (SMCs)
- The middle layer, or tunica media, contains SMCs embedded in a complex extracellular matrix. Arteries affected by obstructive atherosclerosis generally have the structure of muscular arteries
- The arteries often studied in experimental atherosclerosis are elastic arteries, which have clearly demarcated laminae in the tunica media, where layers of elastin lie between strata of SMCs
- The adventitia, the outer layer of arteries, contains mast cells, nerve endings and microvessels

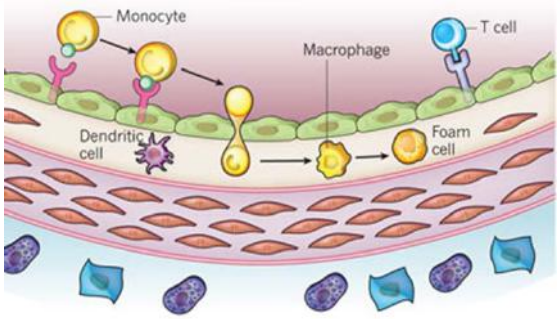
Libby P et al. *Nature*. 2011; **473**(7347): 317-25

Activation of Platelets at Sites of Vascular Injury



Offermanns S. *Circ Res*. 2006; **99**(12): 1293-304

Stages in the Development of Atherosclerotic Lesions

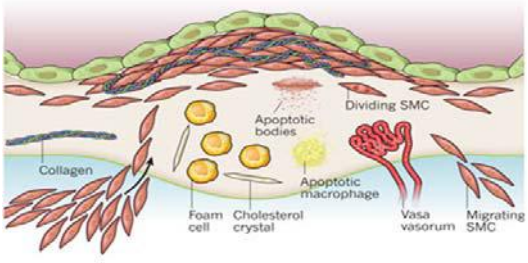


The initial steps of atherosclerosis include:

- Adhesion of blood leukocytes to the activated endothelial monolayer
- Directed migration of the bound leukocytes into the intima
- Maturation of monocytes (the most numerous of the leukocytes recruited) into macrophages, and their uptake of lipid, yielding foam cells

Libby P et al. *Nature*. 2011; **473**(7347): 317-25

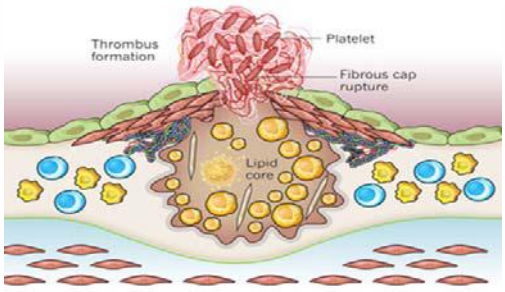
Stages in the development of Atherosclerotic Lesions



- Lesion progression involves the migration of SMCs from the media to the intima, the proliferation of resident intimal SMCs and media-derived SMCs, and the heightened synthesis of extracellular matrix macromolecules such as collagen, elastin and proteoglycans
- Plaque macrophages and SMCs can die in advancing lesions, some by apoptosis
- Extracellular lipid derived from dead and dying cells can accumulate in the central region of a plaque, often denoted the lipid or necrotic core
- Advancing plaques also contain cholesterol crystals and microvessels

Libby P et al. *Nature*. 2011; **473**(7347): 317-25

Stages in the development of Atherosclerotic Lesions



- Thrombosis, the ultimate complication of atherosclerosis, often complicates a physical disruption of the atherosclerotic plaque
- Shown is a fracture of the plaque's fibrous cap, which has enabled blood coagulation components to come into contact with tissue factors in the plaque's interior, triggering the thrombus that extends into the vessel lumen, where it can impede blood flow

Libby P et al. *Nature*. 2011; **473**(7347): 317-25

Blood Pressure

- Hypertension is a multifactorial disorder and signpost that develops because of:
  - Genetic
  - Environmental
  - Anatomical
  - Maladaptive neural, endocrine, humoral, and hemodynamic factors
  - Increased vascular shear stress and stretch due to increased haemodynamics.
- Oxidative stress may contribute to all of these factors
- Production of reactive oxygen species could play an important role in the development of hypertension

Dikalov SI, Ungvari Z. *Am J Physiol Heart Circ Physiol*. 2013; **305**(10): H1417–H1427

Blood Pressure and Arterial Stiffness

- Arterial stiffness and its haemodynamic consequences are established as predictors of adverse cardiovascular outcome
- Arterial stiffness is positively associated with:
  - Systolic hypertension
  - Coronary artery disease
  - Stroke
  - Heart failure

Palatini P et al. *Vasc Health Risk Manag*. 2011; **7**: 725–739

Blood Pressure and Arterial Stiffness

- There is a progressive and graded increase of both systolic and diastolic blood pressure during adolescence and adult life
- After the age of 60-65 years, only the systolic component increases and the diastolic remains stable or even decreases
- While diastolic values only increase until the age of 60–65 years, systolic values constantly increase, although with a different slope, progressively increasing pulse pressure
- The arbitrary cut off of 140 mmHg is often exceeded, leading to a high prevalence of isolated systolic hypertension in the elderly
- A stiff aorta has increased impedance and does not dilate well under pressure, leading stroke volume to impact directly on the arterioles

Palatini P et al. *Vasc Health Risk Manag*. 2011; **7**: 725–739

Blood Pressure

- Factors associated with distribution of BP:

Genetics
Sodium intake (↑ intake leading to ↑ BP in genetically determined sodium-sensitive subjects)
Intake of meat and vegetables (↑ meat/vegetable ratio leading to ↑ BP)
Social stress (↓ stress leading to ↓ BP and to lack of ↑ BP with age)
Income (↓ income leading to ↑ BP)
Culture (↓ culture leading to ↑ BP)
Industrialisation (↑ industrialisation leading to ↑ BP)
Altitude (↑ altitude leading to ↑ BP)
Temperature thermal (↑ temperature leading to ↑ BP)
Water pollution (lead, cadmium)
Air particulate (↑ particulate leading to ↑ BP)

Palatini P et al. *Vasc Health Risk Manag*. 2011; **7**: 725–739

Homocysteine and Cardiovascular Disease

- Observational studies have consistently reported an association between plasma total homocysteine concentrations and the risk of vascular events
- Randomised trials support the hypothesis that mild elevations in homocysteine have a modest effect on cardiovascular risk
- Studies suggest that platelet activation may be a transducer of the effects of high homocysteine in promoting atherothrombosis

Santilli F, Davi G, Patrono C. *Vascul Pharmacol*. 2015. pii: S1537-1891(15)00140-8

Homocysteine and Metabolic Syndrome

- Patients with metabolic syndrome had significantly higher plasma homocysteine levels.
- Increasing homocysteine levels were associated with an increasing prevalence of:
  - Metabolic syndrome
  - Coronary heart disease
  - Cardiovascular disease
- Plasma homocysteine was directly correlated with:
  - Waist circumference
  - Fasting glucose
  - Triglyceride
  - Uric acid
  - Fibrinogen levels
  - Homeostatic model assessment index

Catena C, Colussi G, Nait F et al. *Am J Hypertens*. 2015; **28**(7): 943-50

Homocysteine and Metabolic Syndrome

- Plasma homocysteine was inversely correlated with:
  - Creatinine clearance
  - High-density lipoprotein cholesterol
  - Vitamin B12 level
  - Folate level
- Logistic regression indicated an independent association of homocysteine with cerebro-cardiovascular disease that was independent of the metabolic syndrome

Catena C, Colussi G, Nait F et al. *Am J Hypertens*. 2015; **28**(7): 943-50

MTHFR

- Methyltetrahydrofolate (MTHF) synchronises the activities of the branches of the pathway of methionine biosynthesis by serving as an essential activator of cystathionine gamma-synthase and antagonising the feedback inhibition of this enzyme by S-adenosylmethionine (SAMe)
- Activation is specific for the methylated form of folate and increases with increasing glutamate content
- In *1971*, researchers noted that an excess in the production of Methyl-THF to that of homocysteine would be highly undesirable since the accumulation of THF in methylated form would reduce the amount that is available for purine nucleotide and thymidylate formation

Selhub J et al. *Proc Natl Acad Sci USA*. 1971; **68**(2): 312-4

MTHFR and Homocysteine

- MTHFR is an enzyme involved in the transmethylation pathway, where homocysteine is converted to methionine.
- Circulating folate and homocysteine levels as well as MTHFR genotype, while emerging as major predictors of the risk of vascular events and of the efficacy of folic acid therapy, have also proved to be determinants of an inter-individual variability in the degree of lipid peroxidation and platelet activation, and of the extent of their downregulation by folic acid.
- This may justify a variability and individuality in folate requirements.
- Further dose-finding studies using biochemical endpoints are recommended

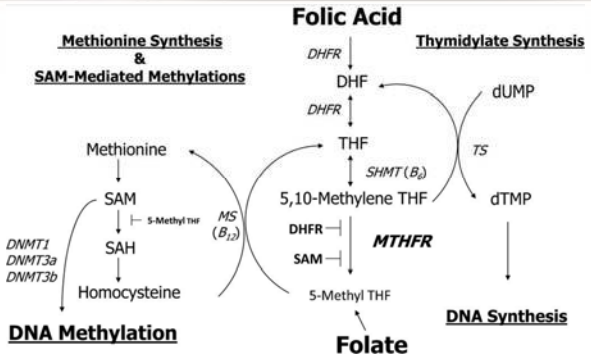
Santilli F, Davi G, Patrono C. *Vascul Pharmacol*. 2015. pii: S1537-1891(15)00140-8

MTHFR: Favourable Evolutionary Factor?

- A recent study has suggested that the MTHFR polymorphism could be a favourable evolutionary factor
- The study found a favourable association with normal renal function of MTHFR polymorphisms
- Notably of MTHFR C677T was present independently of the negative effects of left ventricular hypertrophy, increased intra-renal arterial resistance and hyperparathyroidism
- Results of the study suggested that MTHFR gene polymorphisms could have a protective role on renal function as suggested by their lower frequency among dialysis patients in end-stage renal failure

Trovato FM, Catalano D, Ragusa A et al. *World J Nephrol*. 2015; **4**(1): 127-37

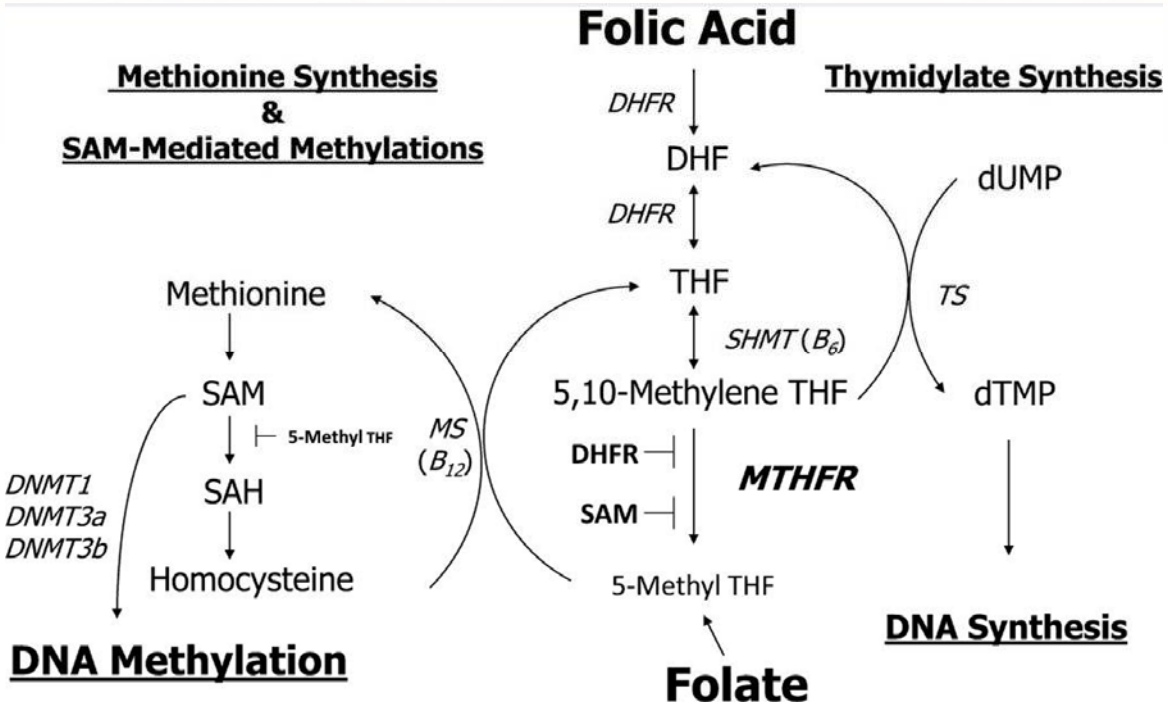
MTHFR and Folic Acid Metabolism



Abbreviations:
- DHF, dihydrofolate;
- DHFR, dihydrofolate reductase;
- DNMT, DNA methyltransferase;
- dTMP, thymidylate;
- dUMP, deoxyuridine monophosphate;
- MS, methionine synthase;
- MTHFR, methylenetetrahydrofolate reductase;
- SAH, S-adenosylhomocysteine;
- SAM, S-adenosylmethionine;
- SHMT, serine hydroxymethyltransferase;
- THF, tetrahydrofolate;
- TS, thymidylate synthase

- The process by which folate/folic acid is used for DNA methylation. The MTHFR 677C→T variant reduces enzyme activity and may help to divert the available methyl groups from the DNA methylation pathway toward the DNA synthesis pathway.
- The pathway is complex and highly regulated. with feedback loops and interactions not shown in the schematic.
- Note: Gene names for enzymes are in italics and cofactors are in parentheses.

MTHFR and Folic Acid Metabolism



Abbreviations:

- DHF, dihydrofolate;
- DHFR, dihydrofolate reductase;
- DNMT, DNA methyltransferase;
- dTMP, thymidylate;
- dUMP, deoxyuridine monophosphate;
- MS, methionine synthase;
- MTHFR, methylenetetrahydrofolate reductase;
- SAH, S-adenosylhomocysteine;
- SAM, S-adenosylmethionine;
- SHMT, serine hydroxymethyltransferase;
- THF, tetrahydrofolate;
- TS, thymidylate synthase

PCOS

- Polycystic ovary syndrome (PCOS) is the most common endocrine disorder amongst women of reproductive age
- Studies have reported a prevalence of approximately 10%
- The following features are more common in women with PCOS than in age-comparable women without PCOS:
  - Insulin resistance
  - Impaired glucose tolerance
  - Type 2 diabetes
  - Obesity
  - Dyslipidaemia

Baldani DP, Skrgatic L, Ougouag R. *Int J Endocrinol*. 2015; 2015: 786362
Randeva HS, Tan BK, Weickert MO et al. *Endocr Rev*. 2012 Oct; **33**(5): 812–841

PCOS

- These metabolic features in patients with PCOS lead to chronic low-grade inflammation and to cardiovascular impairments that potentially elevate the risk of having cardiovascular disease

Baldani DP, Skrgatic L, Ougouag R. *Int J Endocrinol*. 2015; 2015: 786362

Effects of Bereavement on CVD

- Bereavement is associated with increased cardiovascular risk, particularly in surviving spouses and parents
- Inflammatory and thrombotic changes in early bereavement:
  - Compared to non-bereaved participants, those acutely bereaved had a higher neutrophil count, von Willebrand factor antigen, Factor VIII and platelet/granulocyte aggregates
  - Levels of neutrophils, monocytes, eosinophils, platelet count, platelet/monocyte granulocytes and von Willebrand factor were lower in bereaved persons at 6 months compared to acutely

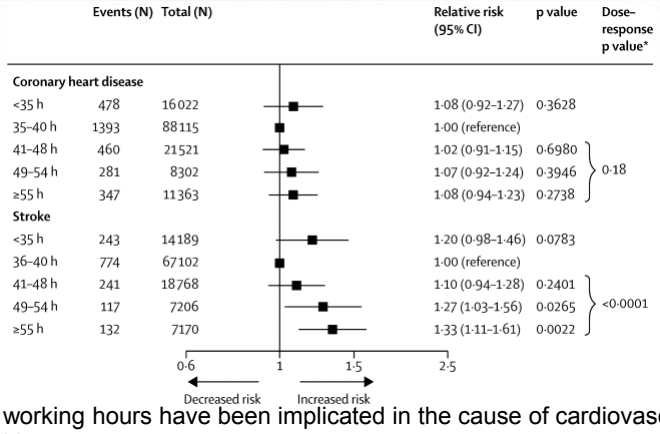
Buckley T, Mihailidou AS, Bartrop R et al. *Heart Lung Circ.* 2011; **20**(2): 91-8

Effects of Bereavement on CVD

- Acutely bereaved (within two weeks) had:
  - Higher 24-hour systolic BP
  - Higher daytime systolic BP
  - Higher daytime systolic load compared to the non-bereaved.
  - By six months the BP of the bereaved tended to be lower than acute measures
- Twenty-four hour heart rate was also higher acutely in bereaved people compared with the reference group
- At six months, the heart rate in the bereaved had fallen to non-bereaved levels

Buckley T, Mihailidou AS, Bartrop R et al. *Heart Lung Circ.* 2011; **20**(2): 91-8

Long Working Hours and Risk of CHD and Stroke



- Long working hours have been implicated in the cause of cardiovascular disease
- Individuals who work 55 hours or more per week have a 1.3 times higher risk of incident stroke than those working standard hours

Kivimäki M, Jokela M, Nyberg ST et al. *Lancet*. 2015; pii: S0140-6736(15)60295-1

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason (↑ = increased, ↓ = decreased)
Glucose (mmol/L)	Fasting: 3.0-5.4 Random: 3.0-7.7	↑ Hyperglycaemia, Diabetes (fasting: > 7.0 mmol/L, 2 hours postprandial > 11.1 mmol/L) ↓ Hypoglycaemia, potential “overdose” of medication
Fasting insulin (mU/L)	<5 during hypo glycaemia; 4-10 after 8 hour fast	↑ levels and ↑ insulin/glucose ratios are found with pancreatic islet beta cell hyperplasia or insulinomas. Non-insulin dependent diabetes and insulin therapy may also give high levels. To identify self-administration of insulin as a cause of hypoglycaemia, C-peptide assay is also required.
HbA1c (%)	< 6.5	≥ 6.5% diagnostic of diabetes *HbA1c levels < 8% indicate “good” control

https://www.rcpa.edu.au/Library/Practising-Pathology/RCPA-Manual/Home

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason (↑ = increased, ↓ = decreased)
CRP (mg/L) hsCRP (mg/L)	< 5 High sensitivity assays (hs-CRP): Low risk: < 1.0 Average risk: 1.0-3.0	↑ acute phase response or active disease in chronic inflammatory disorders. Increased risk of cardiovascular risk in primary prevention populations In patients at risk for myocardial infarction (MI), and without other causes of an acute phase response, the presence of slightly elevated or even high normal hs-CRP indicates a greater risk of MI
Homocysteine (µmol/L)	5-15	↑ risk factor for atherosclerosis and vascular disease. Research found ↑ risk factor for thrombosis in people <40 years of age. ↑ with low levels of folate, B12, B6. MTHFR?

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason (↑ = increased, ↓ = decreased)
Triglycerides (mmol/L)	< 1.7	↑ CVD, diabetes, nephrotic syndrome, hypothyroidism, pancreatitis, alcoholism, oral contraceptive use or corticosteroid medication
Lipoprotein (a) (g/L)	< 0.2	Lipoprotein (a) (Lp (a)) is an independent risk factor for atherosclerosis. ↑ Lp (a) is associated with increased vascular risk. May be indicated in the assessment of a patient with premature coronary or cerebral arterial disease, especially if there is a suggestive family history.

https://www.rcpa.edu.au/Library/Practising-Pathology/RCPA-Manual/Home

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason (↑ = increased, ↓ = decreased)
Total cholesterol (mmol/L)	< 4	↑ risk of coronary artery disease in all age groups. The cause may be primary (familial hypercholesterolaemia and other genetic disorders) or secondary (associated with e.g., biliary obstruction, hypothyroidism, nephrotic syndrome) Levels are reduced for up to 8 weeks with acute illness (e.g., myocardial infarction, acute infection)

https://www.rcpa.edu.au/Library/Practising-Pathology/RCPA-Manual/Home

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason (↑ = increased, ↓ = decreased)
HDL (mmol/L)	**Female:** 1.0-2.2 Target: > 1.0 **Male:** 0.9-2.0 Target: > 1.0	↓ associated with an increased risk of atherosclerotic vascular disease
LDL (mmol/L)	2.0-3.4 Target: < 2.5	↑ associated with an increased risk of atherosclerotic vascular disease. LDL levels are reduced for up to 8 weeks with acute illness (e.g. myocardial infarction, acute infection)

https://www.rcpa.edu.au/Library/Practising-Pathology/RCPA-Manual/Home

Tests for Cardiometabolic Syndrome: a note on HDL sub-fractions

- In a prospective study, HDL2 showed a stronger inverse association with ischemic heart disease risk than did HDL3.
- There are reports that levels of both HDL2 and total HDL were inversely associated with the risk of acute myocardial infarction
- This suggests that these forms of HDL may play a protective role in ischemic heart disease.
- The role of HDL3 remains ambiguous, although small, dense HDL3 has been shown to protect low-density lipoprotein (LDL) from oxidative stress.

Hsieh JY, Chang CT, Huang MT et al. *Anal Chem.* 2013; **85**(23): 11440-8

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason (↑ = increased, ↓ = decreased)
Fibrinogen (g/L)	1.5-4.0	↑ acute phase response, CVD. ↓ reduced production of fibrinogen (liver disease, inherited deficiency), disseminated intravascular coagulation (DIC), fibrinolysis
Von Willebrand Factor (VWF)	50-200% of the level in pooled normal plasma, sometimes expressed as 0.5-2.0 U/mL	Levels of Factor VIII and VWF increase with exercise, stress and endogenous or exogenous oestrogen

https://www.rcpa.edu.au/Library/Practising-Pathology/RCPA-Manual/Home

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason (↑ = increased, ↓ = decreased)
Calcium (mmol/L)	2.10–2.60	↑ Hyperparathyroidism, Vitamin D or Vitamin A toxicity. ↓ Hypoparathyroidism, Renal failure, Osteomalacia or Rickets.
Sodium (mmol/L)	135–145	↑ occurs in a small percentage of patients on diuretic therapy, particularly the elderly. Severe hyperlipidaemia or hyperproteinaemia may cause 'pseudohyponatraemia'. ↓ fluid retention (dilutional hyponatraemia) is seen in renal and cardiac disease
Potassium (mmol/L)	3.5–5.2	↑ acidosis, tissue damage, renal failure, mineralocorticoid deficiency. ↓ in association with loop or thiazide diuretic therapy, vomiting or diarrhoea, alkalosis

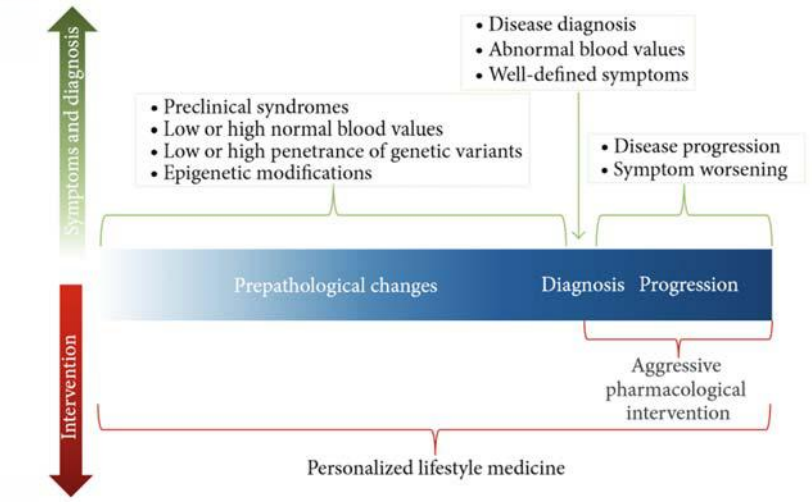
https://www.rcpa.edu.au/Library/Practising-Pathology/RCPA-Manual/Home

Tests for Cardiometabolic Syndrome

Test	Reference range	Reason
MTHFR		Homozygous individuals with the MTHFR C677T mutation have increased homocysteine levels in the presence of low folate stores
Waist:hip ratio	Females: < 0.8 Males: < 0.9	Waist-hip ratio (WHR) has been found to be a dominant risk factor for predicting cardiovascular death in Australia
Waist circumference	Females: > 80 cm Males: > 94 cm	Waist circumference was a better predictor of metabolic syndrome as compared to other obesity indices such as BMI, WHR, and waist height ratio in both men and women Cut-off value of 99.5 cm for men and 91 cm for women has the highest sensitivity and specificity to predict the development of metabolic syndrome

Bener A et al. *J Obes.* 2013; 2013: 269038

The Trajectory of Disease and Role for Personalised Lifestyle Medicine



Minich DM, Bland JS. *Scientific World Journal*. 2013; 2013: 129841

Diet

Diet: Introduction

- Diet plays a fundamental role in:
  - Emotional health
  - Mental health
  - Physical health
  - Quality of life

"The red circles are your red blood cells. The white circles are your white blood cells. The brown circles are donuts. We need to talk."

Diet: Introduction

- Diet plays an essential role in managing cardiometabolic syndrome:
  - Western diet
  - Trans fats
  - Mediterranean diet
  - Fish
  - Chocolate
  - Wine

"The 4 basic food groups are stuff that will make me fat, stuff that will make me sick, stuff that will kill me, and stuff that I'll eat anyway."

Western Diet

- The Western diet pattern reflects a high intake of red and processed meat, refined grains, French fries, and sweets and desserts
- A greater adherence to the Western diet pattern has been linked to a higher risk of:
  - cardiovascular mortality
  - mortality from cancer
  - total mortality

Heidemann C, Schulze MB, Franco OH et al. *Circulation*. 2008; **118**(3): 230-7

Western Diet

- The Western diet pattern has also demonstrated a positive association with the risk of:
  - coronary heart disease
  - stroke
  - type 2 diabetes
  - weight gain
  - elevations of inflammatory and endothelial markers

Heidemann C, Schulze MB, Franco OH et al. *Circulation*. 2008; **118**(3): 230-7

Western Diet and Glycaemic Index

- Greater consumption of refined complex carbohydrates, starches, and sugars (as assessed by glycaemic index or load) was associated with significantly higher risk of:
  - coronary heart disease (36% greater)
  - type 2 diabetes (40% greater)
  - gallbladder disease

Barclay AW, Petocz P, McMillan-Price J et al. *Am J Clin Nutr*. 2008 Mar; 87(3): 627-37

 61

Trans Fats

- The intake of trans fat has been associated with:
  - coronary heart disease
  - sudden death from cardiac causes
  - type 2 diabetes
- In a meta-analysis of prospective cohort studies, each 2% of calories from trans fat was associated with a 23% higher risk of coronary heart disease

Mozaffarian D et al. *N Engl J Med*. 2006; **354**: 1601–1613

 62

Mediterranean Diet and CVD

- Mediterranean dietary pattern is characterised by higher intakes of vegetables, legumes, nuts, fruits, whole grains, fish, and unsaturated fat and lower intakes of red and processed meat
- Greater adherence to the Mediterranean diet has been associated with a lower risk of cardiovascular and total mortality
- In a cohort of 380,296 US men and women, greater versus lower adherence to a Mediterranean dietary pattern was associated with a 22% lower cardiovascular mortality as well as a lower risk of incident CHD and stroke

Mozaffarian D, Benjamin EJ, Go AS et al. *Circulation*. 2015; **131**(4): e29-e322
Heidemann C, Schulze MB, Franco OH et al. *Circulation*. 2008; **118**(3): 230-7

Mediterranean Diet and Type 2 Diabetes

- Adherence to the Mediterranean diet pattern seems to have a protective role on glycaemic control as reflected by reduced:
  - HbA1c level
  - Fasting glucose levels
  - Insulin resistance
  - Peripheral artery disease in patients with type 2 diabetes
  - Mortality
- Decreasing oxidative stress, inflammation, and insulin resistance are all possible mechanism by which Mediterranean diet pauses as a protective dietary pattern

Sleiman D et al. *Front Public Health*. 2015; **3**:69

Fish

- A meta-analysis of 16 prospective cohort studies that included 326,572 generally healthy individuals in Europe, the United States, China, and Japan, fish consumption was associated with significantly lower risk of coronary heart disease mortality
- Compared with no consumption, an estimated 250 mg of long-chain omega-3 fatty acids per day was associated with 35% lower risk of death from coronary heart disease

Mozaffarian D, Benjamin EJ, Go AS et al. *Circulation*. 2015; **131**(4): e29-e322

Fish and Omega-3 content

Name of Fish	Amount of Omega-3 FA (mg/100g)	Amount of Omega-3 FA (mg/150g Serve)	Amount of fish consumption required to Provide 1 g Omega 3 FA per day
Atlantic Salmon	2252	3380	44
Ocean Trout	921	1380	108
Australian Salmon	476	714	210
Rainbow Trout	415	627	241
Red Snapper	357	533	280
Deep Sea Cod	340	510	294
Garfish	327	489	306
John Dory	315	473	317
Barramundi	276	415	362
Coral Trout	270	408	370
Southern Bluefin Tuna	230	345	435

Soltan SS, Gibson RA. *Asia Pac J Clin Nutr*. 2008; **17**(3): 385-90

Chocolate

- Cocoa products containing flavonol have been shown to have an encouraging potential to help prevent cardiometabolic disorders.
- Studies suggest that chocolate consumption has a positive influence on human health via the following:
  - antioxidant
  - antihypertensive
  - anti-inflammatory
  - anti-atherogenic
  - anti-thrombotic
  - influence on insulin sensitivity
  - vascular endothelial function
  - activation of nitric oxide

Buitrago-Lopez A et al. *BMJ*. 2011 Aug; 343: d4488

Red Wine

- Studies suggest that moderate red wine drinkers have a better lipid profile and lower incidence rates of diabetes, hypertension and abdominal obesity
- Compared with non-drinkers, moderate red wine drinkers (≥1 drink/day) were found to have a lower risk of:
  - prevalent metabolic syndrome
  - abnormal waist circumference
  - low HDL-cholesterol concentrations
  - high blood pressure
  - high fasting plasma glucose concentrations

Tresserra-Rimbau A et al. *Br J Nutr.* 2015; **113** Suppl 2:S121-30

Take Home Message…

- Diets such as the Mediterranean diet, high in omega-3 polyunsaturated fatty acids (PUFA), and vegetarian diets have inverse relationships with cardiovascular disease and have the potential to improve glucose control
- A full review of the diet is recommended to assess:
  - nutritional deficiencies
  - allergies and intolerances

Nutrients

Introduction

- Nutrients with potential benefits in cardiometabolic syndrome:
  - Omega-3
  - Coenzyme Q10
  - Lipoic acid
  - Vitamin D
  - N-acetylcysteine
  - Vitamin C
  - Vitamin E
  - Magnesium
  - Zinc
  - Turmeric
  - Resveratrol

Omega-3: Introduction

Omega-3 fatty acids exert pleiotropic cardiometabolic effects with a diverse range of actions:

- Eicosanoid metabolism
- Cell membrane composition
- Cell membrane function
- Anti-inflammatory
- Vascular relaxation
- Improves lipid profile
- Antithrombotic
- Reduce platelet aggregation

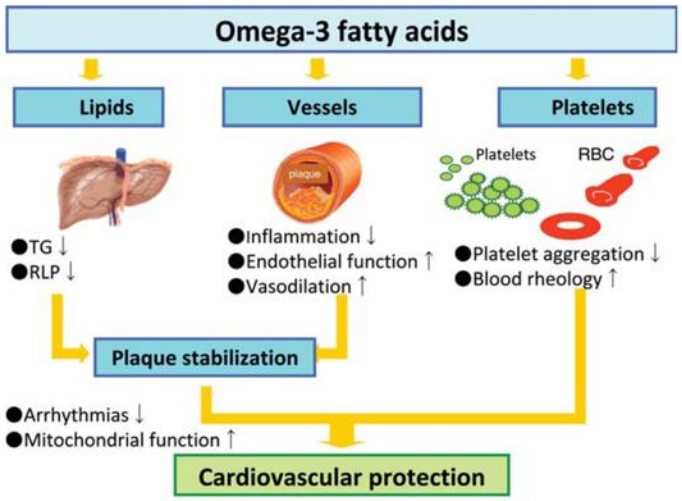
McEwen BJ. *Semin Thromb Hemost*. 2014; **40**(2): 214-26

Omega-3: Introduction

- Omega-3 fatty acids exert pleiotropic cardiometabolic effects with a diverse range of actions:
  - Increase arrhythmic thresholds
  - Reduce blood pressure
  - Improve arterial and endothelial function
  - Prevent the development of plaque and contribute to plaque stabilisation
  - Favourably affect autonomic tone
  - Reduce the risk of arterial atherosclerotic and thrombotic obstruction
  - Increase the myocardial resistance to ischaemia-reperfusion injury

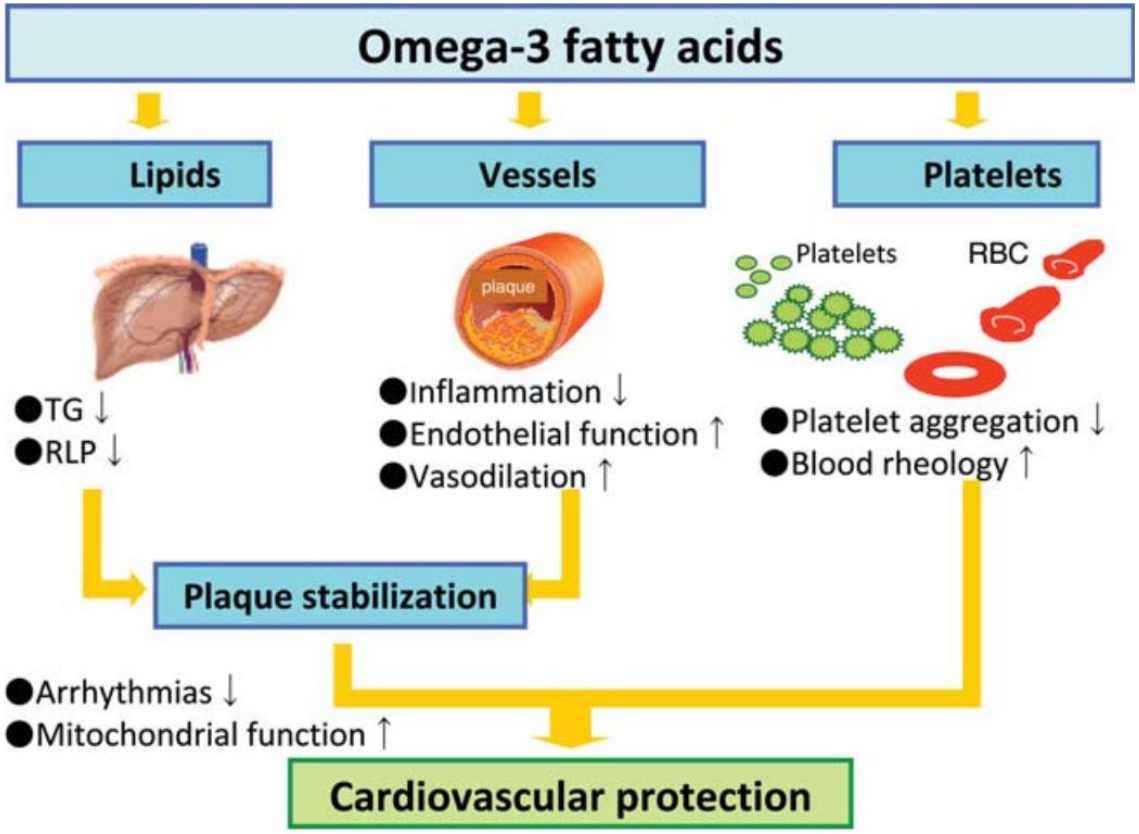
de Lorgeril M, Salen P, Defaye P et al. *BMC Med*. 2013; **11**:5

Omega-3 – Introduction



Kromhout D, Yasuda S, Geleijnse JM et al. *Eur Heart J*. 2012; **33**(4): 436-43

Omega-3 – Introduction



Kromhout D, Yasuda S, Geleijnse JM et al. *Eur Heart J*. 2012; **33**(4): 436-43

Omega-3 and CVD

- There are associations with fish and omega-3 LC-PUFA consumption:
  - Total cardiovascular mortality (approximately 15%-19%)
  - Decreased platelet activation and aggregation
  - Improved lipid profiles, including reduction of triglycerides and very low-density lipoprotein (VLDL)
  - Decreased inflammation
  - Lowered blood pressure

McEwen B, Morel-Kopp MC, Tofler G et al. *Diabetes Educ*. 2010; **36**(4) :565-84

Omega-3 and CVD

- Observational studies show an inverse correlation between fish consumption and cardiovascular disease
- A review of 11 cohort studies involving 116,764 individuals suggested that fish consumption at 40-60 g daily is associated with markedly reduced CHD mortality in high-risk, but not in low-risk populations

Marckmann P, Grønbaek M. *Eur J Clin Nutr*. 1999; **53**(8) :585-90

Omega-3: CVD Risk in Diabetes

- Diets higher in fish and omega-3 LC-PUFA may reduce cardiovascular risk in diabetes by:
  - inhibiting platelet aggregation
  - improving lipid profiles
  - reducing cardiovascular mortality

McEwen B, Morel-Kopp MC, Tofler G et al. *Diabetes Educ*. 2010; **36**(4): 565-84

Omega-3 – Metabolic Syndrome

- Treatment with fish oil in patients with metabolic syndrome for 6 months reduced:
  - serum level of triglycerides
  - LDL-cholesterol
  - hydrogen peroxide
  - malondialdhyde
  - TNF-α
  - IL-6 levels

Al-Gayyar MM, Shams ME, Barakat EA. *Pharm Biol*. 2012; **50**(3): 297-303

Omega-3 and Platelet Function

- Four week study of omega-3 (DHA 520 mg and EPA 120 mg) supplementation:
  - In healthy subjects (n=40), omega-3 PUFA significantly reduced ADP-induced and adrenaline-induced platelet aggregation
  - Reduced P-selectin expression on platelets and platelet-monocyte aggregates after activation with ADP
  - Fewer changes in platelet aggregation and activation found in subjects with CVD (n=16)
  - Significant reduction in slope of arachidonic acid-induced platelet aggregation and increased lag time for U46619-induced platelet aggregation

McEwen BJ et al. *Semin Thromb Hemost.* 2013; **39**(1): 25-32

Omega-3 and Platelet Function

- Four week supplementation of 640 mg omega-3 PUFA reduced measures of platelet aggregation and activation in healthy subjects but effects were less evident in patients with existing CVD
- **Findings support the recommendation that the omega-3 PUFA dose be higher in CVD than among healthy subjects**

McEwen BJ et al. *Semin Thromb Hemost.* 2013; **39**(1): 25-32

Omega-3 and Coagulation

- Four week study of omega-3 (DHA 520 mg and EPA 120 mg) supplementation:
  - In 40 healthy subjects, fibrin generation was significantly reduced, as measured by overall coagulation potential, OHP, velocity of fibrin polymerisation, and significant increase in delay to fibrin generation (p=0.002)
  - The peak of generated thrombin was significantly reduced
  - In 16 subjects with CVD, omega-3 PUFA significantly reduced OHP and significantly increased the lag time to thrombin generation.
  - No other effect on other fibrin and thrombin generation parameters in subjects with CVD

McEwen BJ et al. *Semin Thromb Hemost*. 2015; **41**(3): 315-22

Omega-3 and Coagulation

- Four-week omega-3 PUFA supplementation reduced thrombotic potential in healthy subjects, as shown by reduced fibrin generation and peak thrombin
- There was a greater effect on fibrin generation in healthy subjects compared with those with CVD

McEwen BJ et al. *Semin Thromb Hemost*. 2015; **41**(3): 315-22

Omega-3 and Statins

- Studies have shown that statins increase arachidonic acid, the main omega-6 fatty acid in cell membranes
- This may in turn inhibit the protective effects of omega-3, as omega-6 and omega-3 fatty acids are in competition through various pathways involved in the development and complications of CHD

de Lorgeril M, Salen P, Defaye P et al. *BMC Med*. 2013; **11**:5

Omega-3 - Conclusion

Fish and omega-3 LC-PUFA can be recommended to people with diabetes and included into a diabetes management program

McEwen B, Morel-Kopp MC, Tofler G et al. *Diabetes Educ*. 2010; **36**(4): 565-84

Coenzyme Q10 – Functions

- Coenzyme Q10 functions include:
  - Component of the mitochondrial respiratory chain
  - Endogenous lipid-soluble antioxidant
  - Cellular defence against oxidative damage
  - Regulating membrane fluidity
  - Recycling radical forms of vitamin C and E
  - Protecting membrane phospholipids against peroxidation
  - Protects serum low-density lipoprotein from lipid peroxidation

Ernster L, Dallner G. *Biochim Biophys Acta*. 1995; **1271**(1): 195-204
Dhanasekaran M, Ren J. *Curr Neurovasc Res*. 2005; **2**(5): 447-59

Coenzyme Q10

- CoQ10 is an essential compound found naturally in virtually every cell in humans
- CoQ10 in its reduced form, ubiquinol, inhibits:
  - protein and DNA oxidation
  - lipid peroxidation
- Ubiquinol inhibits the peroxidation of cell membrane lipids and lipoprotein lipids present in circulation, and thereby has a direct anti-atherogenic effect

Garrido-Maraver J, Cordero MD, Oropesa-Avila M et al. *Mol Syndromol*. 2014; **5**(3-4): 187-97

Coenzyme Q10

- Conditions associated with low CoQ10 levels:
  - Cardiovascular disease
  - Diabetes
  - Neurodegenerative disorders
  - Fibromyalgia
  - Cancer
  - Muscular diseases
- The decreased serum CoQ10 levels in patients with type 2 diabetes may be associated with subclinical diabetic cardiomyopathy

Garrido-Maraver J, Cordero MD, Oropesa-Avila M et al. *Mol Syndromol*. 2014; **5**(3-4): 187-97

Coenzyme Q10

- CoQ10 has been found to enhance systolic function in chronic heart failure
- However, the its effectiveness may be reduced with concomitant use of current standard therapies
- CoQ10 may improve beta-cell function and enhance insulin sensitivity, which may reduce insulin requirements for diabetic patients

Garrido-Maraver J, Cordero MD, Oropesa-Avila M et al. *Mol Syndromol*. 2014; **5**(3-4): 187-97
Sander S, Coleman CI, Patel AA et al. *J Card Fail*. 2006; **12**(6): 464-72

Coenzyme Q10

- Studies have shown that CoQ10 supplementation:
  - raised plasma CoQ10 levels
  - improved endothelial function in the brachial artery
  - significantly decreased both systolic and diastolic blood pressure
  - decreased glycosylated haemoglobin (HbA1C)
  - in combination with fenofibrate, markedly improved both endothelial and non-endothelial forearm vasodilation

Garrido-Maraver J, Cordero MD, Oropesa-Avila M et al. *Mol Syndromol*. 2014; **5**(3-4): 187-97

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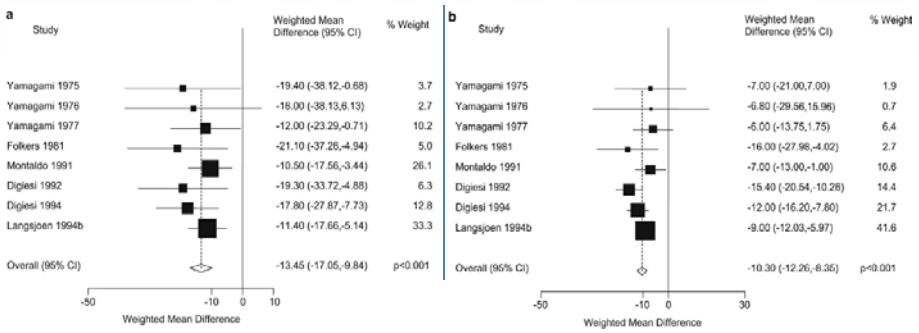
Coenzyme Q10

- An 8 week randomised, double-blind trial investigated the effects of the oral treatment with CoQ10 (60 mg twice daily) in patients known to have essential hypertension with coronary artery disease (CAD) receiving antihypertensive medication
- CoQ10 lowered:
  - Systolic and diastolic blood pressure
  - Fasting and 2-hour plasma insulin
  - Glucose
  - Triglycerides
  - Lipid peroxides
  - Malondialdehyde (biomarker for oxidative stress)
- CoQ10 also increased HDL-cholesterol

(Singh et al. 1999)

 90

Coenzyme Q10 and Blood Pressure

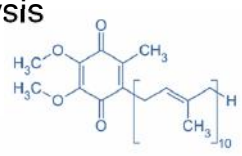


- In open label observational studies:
  - mean systolic blood pressure was decreased by 13.5 mm Hg
  - mean diastolic blood pressure was decreased by 10.3 mm Hg

Rosenfeldt FL, Haas SJ, Krum H et al. *J Hum Hypertens*. 2007; **21**(4): 297-306

Coenzyme Q10 and Statins

- Elevated cholesterol and the associated dyslipidaemia are commonly treated with (HMG-CoA) reductase inhibitors (statins)
- As cholesterol and Coenzyme Q10 synthesis depend on HMG-CoA reductase, both can be blocked
- Depletion in CoQ10 may account for the statin-induced myopathies observed in some patients, the most serious of which is rhabdomyolysis



Garrido-Maraver J, Cordero MD, Oropesa-Avila M et al. *Mol Syndromol*. 2014; **5**(3-4): 187-97

Coenzyme Q10 and Other Medications

- Beta blockers propranolol and metoprolol, and phenothiazines and tricyclic antidepressants have been shown to inhibit CoQ10-dependent enzymes
- CoQ10 acts similarly vitamin K, it may counteract the anticoagulant effects of warfarin

Garrido-Maraver J, Cordero MD, Oropesa-Avila M et al. *Mol Syndromol*. 2014; **5**(3-4): 187-97

Lipoic Acid – Introduction

- Lipoic acid is a naturally occurring dithiol compound synthesised enzymatically in the mitochondrion
- It is a necessary cofactor for mitochondrial α-ketoacid dehydrogenases and serves a critical role in mitochondrial energy metabolism
- Lipoic acid has been used to improve age-associated cardiovascular, cognitive, and neuromuscular deficits

Shay KP, Moreau RF, Smith EJ et al. *Biochim Biophys Acta*. 2009; **1790**(10): 1149-60

Lipoic Acid – Functions

- Lipoic acid functions include:
  - Potent biological antioxidant
  - Scavenges hydroxyl radicals and hypochlorous acid, and terminates singlet oxygen
  - Protects against ischaemic damage
  - Markedly increases intracellular glutathione
  - Increase eNOS activity
  - Activate Phase II detoxification via the transcription factor nuclear factor erythroid 2–related factor 2 (Nrf2)
  - Lower expression of MMP-9 and VCAM-1 through suppression of NFkB
  - Signal transduction

Shay KP, Moreau RF, Smith EJ et al. *Biochim Biophys Acta*. 2009; **1790**(10): 1149-60

Lipoic Acid – Functions

- Lipoic acid may act indirectly to maintain cellular antioxidant status by either inducing the uptake or enhancing the synthesis of endogenous low molecular weight antioxidants or antioxidant enzymes
- Reports show that lipoic acid increases intracellular ascorbate levels
- Lipoic acid is readily converted in various tissues to its reduced form, dihydrolipoic acid, which increases intracellular levels of coenzyme Q10, NADPH, and NADH via increased glutathione availability

Shay KP, Moreau RF, Smith EJ et al. *Biochim Biophys Acta*. 2009; **1790**(10): 1149-60
Sola S, Mir MQ, Cheema FA, Khan-Merchant N et al. *Circulation*. 2005; **111**(3):3 43-8

Lipoic Acid – Potential in Diabetes

- Lipoic acid was also found to enhance expression of the insulin receptor substrate 1 (IRS1) protein in muscle of obese Zucker rats
- Lipoic acid has been proposed to recruit GLUT4 from its storage site in the Golgi to the sarcolemma, so that glucose uptake is stimulated by the local increase in transporter abundance
- Evidence from cell culture experiments supports the involvement of the insulin-signalling cascade in LA-stimulated translocation of GLUT1 and GLUT4
- The cellular distribution of GLUT1 and GLUT4 glucose transporters responded to lipoic acid in a similar fashion as seen with insulin

Shay KP, Moreau RF, Smith EJ et al. *Biochim Biophys Acta*. 2009; **1790**(10): 1149-60

Lipoic Acid – Potential in Diabetes

1. Patients were randomly assigned to oral treatment with 600 mg of lipoic acid t.i.d. (*n*=12) or placebo (*n*=12) for 3 weeks. Neuropathic symptoms (pain, burning, paraesthesiae, and numbness) in the feet were scored at weekly intervals

   Results indicated that after treatment with lipoic acid 600 mg (t.i.d.) for 3 weeks, symptoms and deficits resulting from polyneuropathy in Type 2 diabetic patients may be improved, without causing significant adverse reactions
2. Diabetic patients in the ALADIN II trial were administered intravenous lipoic acid at 600 or 1200 mg/day for 5 days, then oral lipoic acid for 2 years. Lipoic acid improved the indices of neuropathy

Reljanovic M, Reichel G, Rett K et al. *Free Radic Res*. 1999; **31**(3): 171-9
Ruhnau KJ, Meissner HP, Finn JR et al. *Diabet Med*. 1999; **16**(12): 1040-3

Lipoic Acid – Potential in Diabetes

3. A multicentre, randomised, double-blind, placebo-controlled trial investigated the effect of once-daily oral doses of 600 mg (n = 45), 1,200 mg (n = 47), and 1,800 mg of lipoic acid (n = 46) or placebo (n = 43) for 5 weeks after a 1-week placebo run-in period, in 181 diabetic patients with distal symmetric polyneuropathy.

   Significant improvements favouring all three lipoic acid groups was noted for stabbing and burning pain, the Neuropathy Symptoms and Change score.

   Neuropathy Impairment Score sensory function was improved in the lipoic acid 600 mg group compared to placebo

Ziegler D, Ametov A, Barinov A et al. *Diabetes Care*. 2006; **29**(11) :2365-70

Lipoic Acid – Potential in Diabetes

4. The total symptom score (pain, burning, paraesthesia, and numbness) in the feet decreased significantly from baseline to day 19 in the lipoic acid 1,200 mg and lipoic acid 600 mg groups vs. placebo.

   Intravenous treatment with alpha-lipoic acid over 3 weeks was found to be safe and effective in reducing symptoms of diabetic peripheral neuropathy.

   Oral treatment with 800 mg/day for 4 months may improve cardiac autonomic dysfunction in type 2 diabetes.

   No significant adverse events were observed.

Ziegler D, Gries FA. *Diabetes*. 1997; **46** Suppl 2:S62-6

Lipoic Acid – Potential in Diabetes

5. Compared to placebo, 4 weeks of lipoic acid 300 mg:
   - reduced plasma levels of IL-6 by 15%
   - reduced plasma levels of plasminogen activator inhibitor-1 (PAI-1) by 14%
   - Endothelium-dependent flow-mediated vasodilation of the brachial artery was increased by 44% compared with the placebo group

Sola S, Mir MQ, Cheema FA, Khan-Merchant N et al. *Circulation*. 2005; **111**(3): 343-8

Lipoic Acid – Upper Range

- LA supplements up to 2400 mg/day with no reported adverse effects versus placebo
- Oral doses of 1800 mg LA (600 mg t.i.d.) for 6 months did not elicit significant adverse effects compared to placebo

Shay KP, Moreau RF, Smith EJ et al. *Biochim Biophys Acta*. 2009; **1790**(10):1149-60

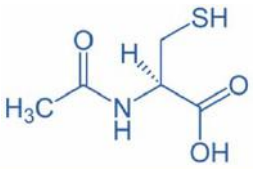
N-Acetylcysteine - Introduction

- N-acetylcysteine (NAC) is an antioxidant and anti-inflammatory agent
- NAC is a glutathione precursor, by entering cells and being hydrolysed to cysteine, it stimulates glutathione synthesis
- NAC increases the level of intracellular reduced glutathione, which is often depleted as a response to increased status of inflammation reaction and oxidative stress

Liu XH, Xu CY, Fan GH. *BMC Cardiovasc Disord*. 2014; **14**: 52

N-Acetylcysteine - Introduction

- NAC has therapeutic value for reducing:
  - endothelial dysfunction
  - inflammation
  - fibrosis
  - cartilage erosion
- NAC improves detoxification of paracetamol



Zafarullah M, Li WQ, Sylvester J et al. *Cell Mol Life Sci*. 2003; **60**(1): 6-20

N-Acetylcysteine - Introduction

- In addition, it may also block renin angiotensin system and/or atrial remodelling via its antioxidant and anti-inflammatory actions
- Oral NAC has been shown to reduce endothelial activation, oxidative stress markers and blood pressure in patients with type 2 diabetes

Treweeke AT, Winterburn TJ, Mackenzie I et al. *Diabetologia*. 2012; **55**(11): 2920–2928
Liu XH, Xu CY, Fan GH. *BMC Cardiovasc Disord*. 2014; **14**: 52

N-Acetylcysteine and Platelets

- Patients were randomised to receive either NAC (1,200 mg) or placebo daily for a total of 8 days
- Platelet–monocyte conjugation was significantly inhibited in patients at 2 hours after administration of NAC
- The inhibition of platelet–monocyte conjugation was maintained (approximately −10 to –15% compared with placebo) on day 7 of once-daily administration of NAC

Treweeke AT, Winterburn TJ, Mackenzie I et al. *Diabetologia*. 2012; **55**(11): 2920–2928

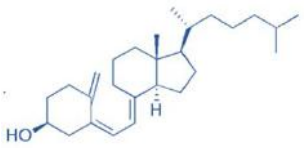
N-Acetylcysteine - Atrial Fibrillation

- NAC could reduce cellular oxidative damage and systematic inflammation during cardiac surgery
- A meta-analysis of 1,026 patients showed that prophylactic use of NAC could reduce the incidence of postoperative atrial fibrillation and all-cause mortality for adult patients undergoing cardiac surgery
- Dosages ranged from 600 mg daily to 1,200 mg twice daily

Liu XH, Xu CY, Fan GH. *BMC Cardiovasc Disord*. 2014; **14**: 52

Vitamin D – Introduction

- Epidemiological studies have linked vitamin D deficiency with:
  - Cardiovascular disease
  - Chronic illnesses
  - Autoimmune disease
  - Cancer
  - Depression
  - Dementia
  - Infectious diseases
  - Musculoskeletal decline



Holick MF. Vitamin D deficiency. *N Engl J Med*. 2007; **357**(3): 266-81

Vitamin D – Introduction

- A meta-analysis demonstrated a linear, inverse association between circulating 25(OH)-vitamin D in the range of 20-60 nmol/L and risk of CVD

Wang L, Song Y, Manson JE et al. *Circ Cardiovasc Qual Outcomes*. 2012; **5**(6): 819–829

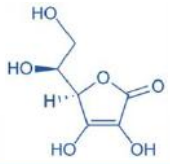
Vitamin D and Metabolic Syndrome

- Vitamin D has pleiotropic effects (having multiple effects from a single gene) that may favourably influence cardiovascular health through multiple mechanisms:
  - down-regulation of the renin-angiotensin system
  - enhancement in insulin secretion and insulin sensitivity
  - protection against angiogenesis
  - modulation of inflammatory processes

Wang L, Song Y, Manson JE et al. *Circ Cardiovasc Qual Outcomes*. 2012; **5**(6): 819–829

Vitamin C

- Vitamin C is present in two biological important forms:
  - the reduced form, ascorbic acid
  - the oxidised form, dehydroascorbic acid
- Both forms are transported intracellularly
- Once inside the cells, dehydroascorbic acid is rapidly reduced to ascorbic acid



Rodrigo R, Libuy M, Feliu F et al. *Biomed Res Int*. 2013; 2013: 437613

Vitamin C

- Vitamin C is a reducing agent that serves as a one-electron donor, generating semidehydroascorbate
- When it acts as an antioxidant or enzyme cofactor, it becomes oxidised to dehydroascorbic acid
- Ascorbate counteracts and prevents the oxidation of lipids, proteins, and DNA, subsequently protecting their structure and biological function
- Together with glutathione, ascorbic acid constitutes a primary line of defence against ROS

Rodrigo R, Libuy M, Feliu F et al. *Biomed Res Int*. 2013; 2013: 437613

Vitamin C

- Ascorbate in aqueous compartments can recycle α-tocopherol in membranes by reducing the α-tocopheroxyl radical back to α-tocopherol
- Ascorbate has been shown to recycle α-tocopherol in lipid bilayers and erythrocytes
- The antioxidant effect of ascorbic acid is not limited to its ability to scavenge ROS
- Ascorbate can reduce ROS production through downregulation of NADPH oxidase
- The therapeutic potential of vitamin C in acute myocardial infarction is via reduction of NADPH oxidase, the major source of ROS production

Rodrigo R, Libuy M, Feliu F et al. *Biomed Res Int*. 2013; 2013: 437613

Vitamin C

- Vitamin C also suppresses NF-kB activation
- Vitamin C prevents the oxidation of tetrahydrobiopterin, a cofactor of nitric oxide synthase that is highly sensitive to oxidation
- When tetrahydrobiopterin is oxidised, eNOS activity becomes uncoupled, resulting in the production of superoxide instead of nitric oxide, enhancing the oxidative damage

Rodrigo R, Libuy M, Feliu F et al. *Biomed Res Int*. 2013; 2013: 437613

Vitamin E

- Vitamin E, mainly α-tocopherol, is the major peroxyl radical scavenger in biological lipid-phases such as membranes or LDL
- The antioxidant action is via its ability to act chemically as a lipid based free radical chain-breaking molecule, thereby inhibiting lipid peroxidation through its own conversion into an oxidised product, α-tocopheroxyl radical
- α-tocopherol can be restored by reduction of the α-tocopheroxyl radical with redox-active reagents like vitamin C or ubiquinol

Vitamin E

- The antioxidant effect of vitamin E is not limited to the role of lipid phase ROS scavenger. Vitamin E also has the following functions:
  - increasing glutathione peroxidase activity
  - synergism with other antioxidant molecules
  - reducing ROS production via downregulating NADPH oxidase
  - anti-inflammatory effects:
    - by inhibiting the transcriptional activity of NF-kB, thereby reducing the expression of pro-inflammatory genes
    - reducing the release of pro-inflammatory cytokines
  - reducing platelet aggregation

Rodrigo R, Libuy M, Feliu F et al. *Biomed Res Int*. 2013; 2013: 437613

Vitamin C and E

- Vitamins C and E exert their individual biochemical effects in water or lipid phases, respectively
- They can interact with each other at the level of interphases, developing the synergistic effects of restoring α-tocopherol from α-tocopheroxyl radical

Rodrigo R, Libuy M, Feliu F et al. *Biomed Res Int*. 2013; 2013: 437613

Vitamin C and E

- A randomised, double-blind, placebo-controlled study measured 1000 mg/12 hour infusion of vitamin C followed by 1200 mg/day orally and vitamin E (600 mg/day) for 30 days
- Results suggested the positive influence of C and E on the clinical outcome of patients with acute myocardial infarction
- Supplementation showed less frequent:
  - in-hospital cardiac mortality
  - nonfatal new myocardial infarction
  - ventricular tachycardia
  - ventricular fibrillation
  - asystole or shock
  - pulmonary oedema

Jaxa-Chamiec T et al. *Kardiol Pol.* 2005; **62**(4): 344-50

Magnesium – Introduction

- Magnesium is an essential trace mineral that plays a key role in all energy-dependent transport systems, glycolysis and oxidative energy metabolism
- Low serum magnesium levels have been associated with risk factors of metabolic syndrome:
  - hyperglycaemia
  - hypertension
  - hypertriglyceridaemia
  - insulin resistance

Ju SY, Choi WS, Ock SM et al. *Nutrients*. 2014; **6**(12): 6005-19

Magnesium – Introduction

- Additionally, waist circumference is independently associated with hypomagnesaemia
- Findings suggest that low magnesium concentration may contribute to the pathogenesis of coronary atherosclerosis or acute thrombosis

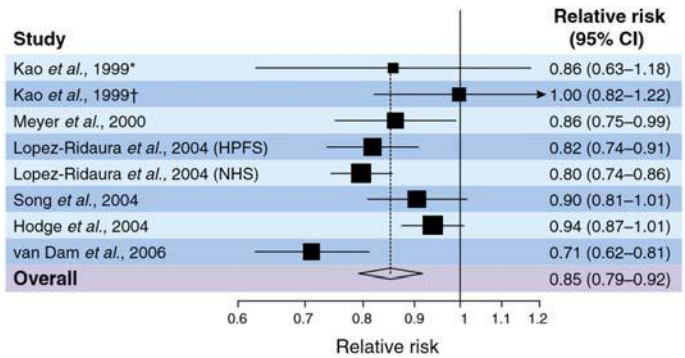
Ju SY, Choi WS, Ock SM et al. *Nutrients*. 2014; **6**(12): 6005-19
Liao F, Folson AR, Brancati FL et al. *Am Heart J*. 1998; **136**(3): 480-90

Magnesium and CVD

- Oral magnesium (365 mg) for 6 months in coronary artery disease patients resulted in significant improvement in brachial artery endothelial function and exercise tolerance
- These results suggest a potential mechanism by which magnesium could beneficially alter outcomes in patients with coronary artery disease

Shechter M, Sharir M, Labrador MJ et al. *Circulation*. 2000; **102**(19): 2353-8

Magnesium Intake and Incidence of Type 2 Diabetes



Association between magnesium intake (for a 100 mg/day increase) and incidence of Type 2 diabetes. In the various trials, the study-specific relative risk was assessed (squares). The diamond represents the overall relative risk.

Reffelmann T, Ittermann T, Dorr M et al. *Atherosclerosis*. 2011; **219**(1): 280-4

Magnesium and Metabolic Syndrome

- In a randomised double-blind placebo-controlled trial, individuals received either 382 mg of magnesium or placebo for 4 months
- Compared to placebo, patients in the magnesium group had significant reductions in:
  - systolic and diastolic blood pressures
  - HOMA-IR index
  - fasting glucose
  - triglyceride levels

Rodríguez-Moran M, Guerrero-Romero F. *Arch Med Res*. 2014; **45**(5): 388-93

Magnesium and Metabolic Syndrome

- A meta-analysis of observational studies showed a statistically significant inverse association between dietary magnesium intake and the risk of metabolic syndrome based on cross-sectional studies and prospective cohort studies published between 2005 and 2013
- The overall estimate indicated a 12% reduction in the risk of metabolic syndrome for an increment in magnesium intake of 150 mg/day

Ju SY, Choi WS, Ock SM et al. *Nutrients*. 2014; **6**(12): 6005-19

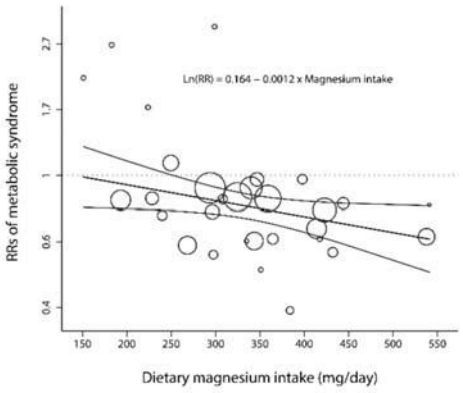
Magnesium and Metabolic Syndrome

- Magnesium intake of 150 mg/day is approximately equivalent to:
  - 56 g of dry roasted almonds
  - one cup of cooked spinach
  - 1.5 cup of beans
  - two cups of brown, long-grained cooked rice
  - three medium baked potatoes with skin
  - five medium bananas
  - six tablespoons of peanut butter per day

Ju SY, Choi WS, Ock SM et al. *Nutrients*. 2014; **6**(12): 6005-19

Association Between Risk of Metabolic Syndrome and Dietary Mg Intake

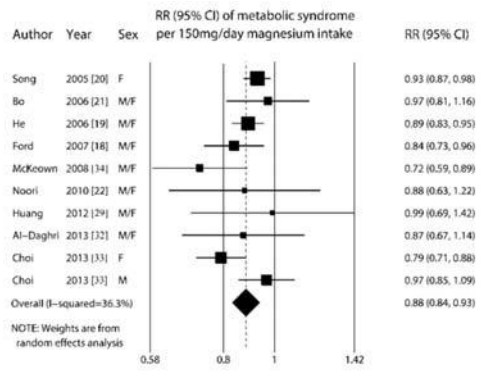
- Dose-response meta-regression
- The levels of magnesium intake (mg/day) were modelled using a linear trend with random-effects meta-regression models
- The solid black line represents the weighted regression line based on variance-weighted least squares
- The grey line shows the 95% CI around the regression line
- The circles indicate RRs in each study. The circle size is proportional to the precision of the RR



Ju SY, Choi WS, Ock SM et al. *Nutrients*. 2014; **6**(12): 6005-19

Magnesium and Metabolic Syndrome

- Forest plots of the risks ratios (RRs) of metabolic syndrome per 150 mg/day increment in dietary magnesium intake (n = 30,092) using a random-effects analysis
- The squares represent study-specific RR (the square sizes are proportional to the weight of each study in the overall estimate)
- The horizontal lines represent 95% confidence intervals (CIs)
- The diamond represents the overall RR estimate with the 95% CI



Ju SY, Choi WS, Ock SM et al. *Nutrients*. 2014; **6**(12): 6005-19

Zinc – Introduction

- Second most common trace metal in the body and is an essential micronutrient
- Plays crucial roles in the synthesis, storage and secretion of insulin and in the actions of insulin on carbohydrate metabolism – zinc possesses an insulinomimetic effect
- Plays vital roles:
  - in growth and development
  - as a cofactor for metalloenzymes in antioxidant defence systems (superoxide dismutase, catalase, and glutathione peroxidase)
  - reduces inflammatory cytokine production via regulation of a zinc-finger protein
  - inhibition of NF-κB activation
  - decreases ROS generation and induces metallothionein, which decreases the OH burden

Seo JA, Song SW, Han K et al. *PLoS One*. 2014; **9**(8): e105990

Zinc and Metabolic Syndrome

- The results of this study showed associations between serum zinc levels and certain metabolic syndrome components
- There was no significant association found between serum zinc levels and the prevalence of metabolic syndrome in either men or women
- In both men and women, significant negative correlations were observed between serum zinc levels and fasting glucose and the HOMA-IR index
- There were differences in serum zinc levels according to the number of metabolic syndrome components in women:
  - serum zinc levels decreased as the number of metabolic syndrome components increased
  - inverse correlations between serum zinc levels and systolic blood pressure and insulin levels
  - low serum zinc levels showed a greater prevalence than the highest serum zinc levels among almost every metabolic syndrome phenotype

Seo JA, Song SW, Han K et al. *PLoS One*. 2014; **9**(8): e105990

Zinc and Metabolic Syndrome

- A randomised, double-blind, placebo-controlled trial investigated the effect of 50 mg zinc vs placebo among 52 women diagnosed with PCOS and aged 18-40 years old
- After 8 weeks of intervention, subjects who received zinc 50 mg (elemental) supplement had reduced:
  - fasting plasma glucose
  - serum insulin levels
  - homeostasis model of assessment-insulin resistance (HOMA-IR)
  - homeostatic model assessment-Beta cell function (HOMA-B)
  - serum triglycerides
  - VLDL-cholesterol
- In addition, zinc supplemented women had significantly increased serum zinc levels compared with placebo

Jamilian M, Foroozanfard F, Bahmani F et al. *Biol Trace Elem Res*. 2015. PMID: 26315303.

Turmeric – Introduction

- Turmeric/Curcumin plays a role in decreasing:
  - platelet aggregation
  - oxidative processes
  - inflammatory cytokine production
  - NF-kB
  - pro-inflammatory interleukins (IL-1, -2, -6, -8, -12)
  - cholesterol levels
  - monocyte chemoattractant protein-1 (MCP-1)
- In addition, curcumin improves wound healing and modulates signalling pathways
- Curcumin also down-regulates the activity of cyclooxygenase-2 (COX-2) and lipoxygenase

Ghorbani Z et al. *Int J Endocrinol Metab*. 2014; **12**(4): e18081

Turmeric and Metabolic Syndrome

- Curcumin has the potential for reducing metabolic syndrome via:
  - reduction in blood glucose and HbA1c levels
  - reduction in hepatic glucose production and glycogen synthesis
  - stimulation of glucose uptake by increasing GLUT4, GLUT2 and GLUT3 gene expressions
  - increasing activation of AMP kinase (central regulator of cellular metabolism and homeostasis)
  - promoting PPAR γ ligand-binding activity (regulates cellular differentiation, fatty acid storage and glucose metabolism)
  - suppressing hyperglycaemia-induced inflammatory state
  - stimulation of insulin secretion from pancreatic tissues

Ghorbani Z et al. *Int J Endocrinol Metab*. 2014; **12**(4) :e18081

Turmeric and Metabolic Syndrome

- Curcumin has the potential for reducing metabolic syndrome via:
  - improvement in pancreatic cell function – increasing phosphorylation of AKT (protein kinase B; PKB)
    - a central player in the signal transduction pathways activated in response to growth factors or insulin
    - contributes to several cellular functions including nutrient metabolism, cell growth, and transcriptional regulation
  - reduction of insulin resistance
- Glucose lowering effect of turmeric and curcumin has been observed in human clinical trials conducted on diabetic and pre-diabetic patients
- However, no effect was seen in patients with normal baseline levels of blood sugar

Ghorbani Z et al. *Int J Endocrinol Metab*. 2014; **12**(4): e18081

Resveratrol and Metabolic Syndrome

- In a double-blind, randomised, placebo-controlled trial, 60 subjects with non-alcoholic fatty liver disease were given 2 placebo capsules or 2 x 150mg resveratrol capsules twice daily for three months
- Compared with the placebo, resveratrol significantly decreased:
  - glucose levels
  - low-density lipoprotein cholesterol
  - total cholesterol
  - homeostasis model assessment insulin resistance index (HOMA-IR)
  - levels of TNF-α
  - alanine aminotransferase
  - aspartate aminotransferase
- An elevation of adiponectin level was observed

Chen S et al. *Dig Liver Dis*. 2015; **47**(3): 226-32

Folate

- The effect of folate on methylation and on the metabolism of methionine has been known since the 1950's

Dinning J, Keith CK, Day PL. *J Biol Chem*. 1951; **189**(2): 515-20

Additional Nutrients for Cardiometabolic Health

These nutrients are active and synergistic nutrients that play a beneficial in the management of cardiometabolic syndrome.

- Folate/Folinic acid
- B12
- B6
- Biotin
- Chromium
- Manganese
- Potassium
- Selenium
- Carnitine
- Glutamine
- Taurine

Take home message…

- Synergism plays an important role in reducing the risk factors of cardiometabolic syndrome
- Review nutrients for synergism and antagonism
- Dosage depends on the severity of the condition

Lifestyle

Sleep

- Short sleep may result in hormonal and metabolic changes contributing to weight gain and obesity
- Lower average sleep duration is consistently linked to greater adiposity in both children and adults
- Short-term trials demonstrate effects of insufficient sleep on hunger, food choices, and leptin/ghrelin concentrations

Magee L, Hale L. *Sleep Med Rev.* 2012; **16**(3): 231-41

Sleep

- An association between sleep deprivation and thermoregulation decreases the available energy expenditure and is related to increased food intake
- Short sleep duration has been associated with fatigue and reduced physical activity, a source of reduced caloric expenditure
- Being awake longer gives one more opportunity to eat, coupled with the fact that sleep deprivation may affect choices about food quality

Magee L, Hale L. *Sleep Med Rev.* 2012; **16**(3): 231-41

Physical Activity

- A total of 120 to 150 minutes per week of moderate-intensity activity, compared with none, can reduce the risk of developing metabolic syndrome
- Self-reported low lifetime recreational activity has been associated with increased peripheral artery disease
- Exercise for weight loss, without dietary interventions, was associated with significant reductions in:
  - DBP (–2 mm Hg; 95% CI, –4 to –1 mm Hg)
  - triglycerides (–0.2 mmol/L; 95% CI, –0.3 to –0.1 mmol/L)
  - fasting glucose (–0.2 mmol/L; 95% CI, –0.3 to –0.1 mmol/L)

Mozaffarian D, Benjamin EJ, Go AS et al. *Circulation*. 2015; **131**(4): e29-e322

Stress Management

- Type D (Distressed) personality has been related to an increased prevalence of metabolic syndrome and a unhealthy lifestyle
- Type D personality has been associated with an increased risk of adverse cardiac events in patients with a cardiovascular condition
- Both behavioural (e.g. poor consultation behaviour) and biological (e.g. cortisol hyperactivity, cardiovascular hyper-reactivity, immune factors) mechanisms have been suggested

Mommersteeg PM, Kupper N, Denollet J.. *BMC Public Health*. 2010; **10**:714

Stress Management

- People with Type D personality:
  - made poorer lifestyle choices
  - adhered less to the physical activity
  - had a less varied diet
  - were less likely to restrict their fat intake
- Men were less likely to eat a varied diet and to restrict the amount of salt and fats in their diet
- Individuals with a Type D personality have the tendency to experience increased negative emotions and inhibit these emotions in social situations, because of fear of rejection or disapproval

Mommersteeg PM, Kupper N, Denollet J.. *BMC Public Health.* 2010; **10**:714

Take home message…

- Quality sleep, regular physical activity and stress management are recommended as mainstays in clinical practice
- Graded physical activity may be required so there is no extra undue pressure on the body systems. Basic example is a daily walk
- Stress management can include meditation, massage, tai chi, etc.
- Find out what the patient likes doing and then having them do it regularly

Case studies

Case studies: Male 45

- Male 45
- BP 155/95, Pulse 82, BMI 30
- Stress 7/10
- Takeaway fast food diet
- Abdominal obesity
- Fatigue
- Radiating back pain
- Poor sleep
- Blood tests: elevated cholesterol and triglyceride levels, fasting blood glucose, HbA1c, CRP, and homocysteine.
- Family History: Father MI 50, Mother DVT 48

Case studies: Male 45

- Diet
- Foods
- Nutrients
- Lifestyle

Case studies: Female 48

- Female 48
- BP 140/95, Pulse 78, BMI 25.5
- Stress 7/10, Anxiety
- Western diet and an “apple a day”
- History of PCOS
- Fatigue
- Hot flushes
- Blood tests: elevated cholesterol, HbA1c
- Family History: Father MI 58, Mother PCOS

Case studies: Female 48

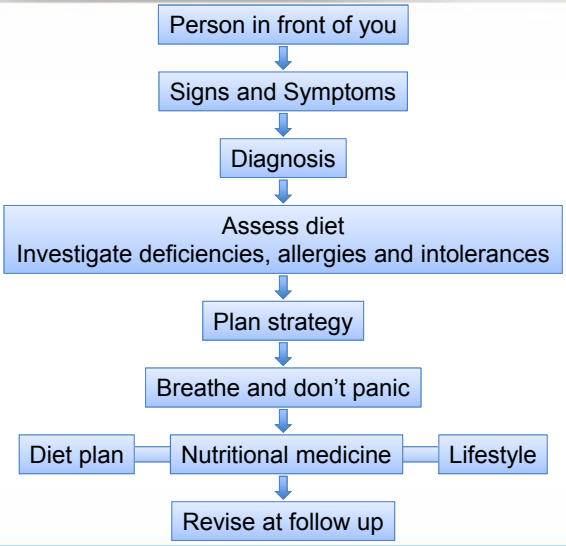
- Diet
- Foods
- Nutrients
- Lifestyle

Final take home message…

Final take home message…

- Nutritional medicine plays an essential role in the management of cardiometabolic syndrome, cardiovascular disease and diabetes
- Lifestyle factors also play major roles in the management of cardiovascular disease

Final take home message…



Final, final take home message…

- Although cardiometabolic syndrome is a complex condition, it can be managed
- With confidence you can make a big difference in someone's life
- Forge ahead…

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INTEGRIA HEALTHCARE SYMPOSIUM 2015

Clinical Pearls + Practitioner Perspectives

An Experience-Based Approach to Patient Care

24+25 OCTOBER
MELBOURNE

Clinical Pearl:
('klɪnɪkəl pûrl)

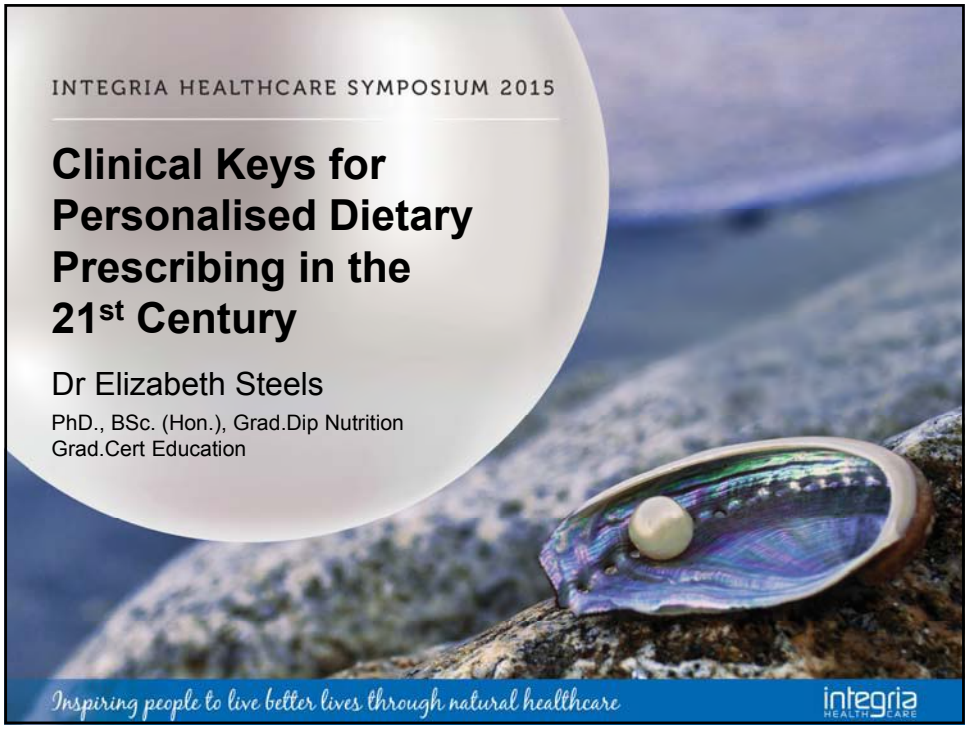
1. A straightforward and meaningful piece of clinical advice
2. Clinically relevant information based on experience

Session 6

Clinical Keys for Personalised Dietary Prescribing in the 21st Century

DAY 2

Dr Elizabeth Steels
PhD, BSc. (Hon.), Grad.Dip Nutrition, Grad.Cert Education



Health in the 21st Century

Food Gurus

What we eat
Where we eat
How we eat

Neal Perry
Donna Hay
Matt Moran
Margaret Beer
Julie Goodwin
Curtis Stone
Adriano Zumba
Kylie Kwong

"Very basically, the food and drink sector has been one of the most reliable sectors of the book market in recent years."

"With their powerful restaurants, high-profile media roles and impeccable palates, they are the state's - and the nation's - most influential voices in food. And not all of them are chefs. In fact, to capture the hearts and minds of increasingly discerning diners, self-proclaimed foodies and home cooks, you don't even need to own a restaurant or be a chef".

Philip Stone, The Bookseller

Health in the 21st Century

- **Predicted Top Selling Cook Books 2015**
  - Nigellissima: Instant Italian Inspiration
    Nigella Lawson
  - Jamie's 15-Minute Meals
    Jamie Oliver
  - Jerusalem
    Yotam Ottolenghi and Sami Tamimi
  - The Kitchen Diaries II
    Nigel Slater
  - Gordon Ramsay's 'Ultimate Cookery Course'
    Gordon Ramsay

Health in the 21st Century

3D food printing is more than just a novelty,
it is the future of food…

Food printing
"is something that will become a part of the culinary fabric."

Health in the 21st Century

Technological Age also brings:

- Stem cell therapy
- Gene-related therapies
- Nutrigenomics

Health in the 21st Century

- According to WHO:
  - Healthier mothers and babies
  - More live births
  - Decline in deaths from heart disease
  - Safer and healthier foods
  - Fortification of foods

Health in the 21st Century

…But 21st Century children have:

- Low birth weight
- Rising rates of obesity and diabetes
- Childhood asthma and other allergies
- A range of developmental disorders (i.e. autism)
- Mental health issues (i.e. depression, anxiety, behavioral disturbance)
- Learning disabilities, aggressive behavior, violence
- Children living in rural/lower socioeconomic groups are particularly at risk

 12

Health in the 21st Century

- **PREVENTATIVE STRATEGIES** are **NOT** a priority!
  - Preventive initiatives do not reach those at risk
  - People with funds can have more diagnostic tests, can afford good food, do exercise classes, choose their occupation and manage their lives better
  - Medical errors in Australia cost over $1 billion (possibly $2 billion) annually
  - The ‘Quality in Australian Health Care’ study found that about half of these errors were potentially preventable

 13

Health in the 21st Century

… Including real food in the diet DOES NOT counteract the adverse effects of the unhealthy foods.

Introduction

- **Part 1:** From the roots of naturopathic medicine, specific dietary prescriptions should be based on **composition of foods tailored to a particular body composition and metabolism**

12 Naturopathic Principles

Traditional, supported by science.

Introduction

- **Part 2:** From the era of modern science, understanding food compositions that are best for our genetic profile, as well of the **effect of dietary intervention on gene expression** will also be essential knowledge to 21st century nutritional medicine

Nutrigenomics

Science supports naturopathic principles.

Introduction

Understanding the interrelationships among:

- Diets and individual chemicals; and
- Effects of and on our genome…

…WILL become a fundamental part of nutritional practice

Introduction

- It will also help identify those who will benefit most from, or be placed at risk by intervention strategies; including particular diets, exercise regimes, supplement and drug interventions to prevent and treat disease

12 Naturopathic Principles

12 Naturopathic Principles apply to ALL 'diets'…

Naturopathic Principle 1

- All FOODS, in their natural form, have a composition that contains macro- and micro-nutrients
- They provide energy, nutrients and the processing aids (vitamins and minerals) in balance
- Majority of foods, if not all, should be in the natural form
- All research supports this

EAT WHOLE FOODS




Naturopathic Principle 2

- Processing of FOODS alters this nutritional balance
- Simple processing (drying) – When eating dried fruits, limit amounts and balance with glass of water
- Simple processing also alters the effect of the original food on GIT (digestion and absorption) and metabolism (metabolic differences between apples, whole, pureed and juiced… no longer an apple!)

REDUCE PROCESSED FOOD



An Apple (or Two) a Day?

- Apples contain Fiber 2-3% (cellulose, hemicullose, pectin) and polyphenols and bioactive components such as dihydrocholcones, flavonols, hydroxycimmates, flavanols (catechin, proanthrocyanidins)
- Epidemiology studies:
  - Apples reduce cardiovascular disease, diabetes
- Interventional studies
  - Support lipid metabolism, weight, vascular function, reduce inflammation, lowered ischemic heart disease mortality, thrombotic stroke, reduce risk of CHD
- Note: Processing to clear juice removes most polyphenols and fiber

Koutsos A, Tuohy KM, Lovegrove JA. *Nutrients*. 2015; **7**: 3959-3998

An Apple (or Two) a Day?

- Apple pectin lowers cholesterol, inhibiting cholesterol uptake, support bile acid production and bind bile acids, supports glucose metabolism. It is a geling agent and impacts on transit time, gastric emptying and nutrient absorption, it is resistance to gastric and enzymatic enzymes and is fermented into SCFA

Koutsos A, Tuohy KM, Lovegrove JA. *Nutrients*. 2015; **7**: 3959-3998

An Apple (or Two) a Day?

- Pectin and the larger polyphenols reach the colon and are fermented by colonic microbes producing metabolites that influence gut microbiota and systemic effects after absorption including butyric acid
- Dietary Prebiotic – a selectively fermented ingredient that results in specific changes in the composition and/or activity of the gastrointestinal microbiota thus conferring benefits to the host health

Koutsos A, Tuohy KM, Lovegrove JA. *Nutrients*. 2015; **7**: 3959-3998

Table 2. Effects of apples and apple components on gut microbiota composition and activity.

Type of Study	Duration-Diets-Daily Dose	Techniques Used	Results	Author
Animal (Wistar rats)	6 weeks, 10 rats for each group: Control diet or 5% apple pomace (AP) 1B-juice colloids extract (54.3% soluble and 2.6% insoluble fiber) or 5% AP 4B-juice colloids extract, rich in soluble fiber (78.3% soluble and 1.8% insoluble) or 5% alcohol AP extract, rich in insoluble fiber (22.9% soluble and 73.3% insoluble)	FISH (caecal) Plate count (feaces)	B-juice AP extracts: Total SCFA, acetate and propionate: ↑, pH: ↓ *Bacteroidaceae*: ↑ (fieces) Alcohol AP extract: Total SCFA and butyrate: ↑, pH: ↓ *Bacteroidaceae*: ↑ (faeces) *Eubacterium rectale*: ↑ (caecal)	Sembries *et al.* (2003) [24]
Animal (Wistar rats)	4 weeks, 12 rats for each group: Control diet or Extraction juice from apple pomace	Plate count	Total SCFA and acetate: ↑, pH: ↓ *Lactobacillus*: ↑, *Bifidobacterium*: ↑ Primary bile acids and neutral sterols: ↑ Secondary bile acids: ↓	Sembries *et al.*, 2006 [25]
Animal (Fischer rats)	4 weeks, 8 rats for each group: Control diet or 10 g apple or 7% apple pectin	qPCR	Apple: Butyrate:↑, pH: ↓, *Bacteroides spp*: ↓ Apple pectin: Butyrate:↑, pH: ↓, *Bacteroides spp*: ↓, *Clostridium coccoides*: ↑	Licht *et al.*, 2010 [26]
Animal *ex vivo* (mice)	Granny Smith apple fermented *in vitro* from faeces from diet induced lean (control) and obese mice.	qPCR	*Firmicutes*, *Bacteroidetes*, *Enterococcus*, *Enterobacteriaceae*, *Escherichia coli* and *Bifidobacterium* abundances from obese mice tended to be similar to lean mice after apple fermentation.	Condezo-Hoyos *et al.*, 2014 [118]
Human	2 weeks, 8 subjects: 2 apples	Plate count	Bifidobacteria: ↑ Clostridia: ↓, *Enterobacteriaceae*: ↓	Shinohara *et al.*, 2010 [27]
Human Randomized, single blinded, controlled, crossover	4 weeks, 23 subjects: Control: period of restricted diet or 550 g whole apples or 22 g apple pomace or 500 ml clear apple juice or 500 ml cloudy apple juice	qPCR	No changes in bacteria composition. pH: ↓	Ravn-Haren *et al.*, 2012 [15]

↑: significant increase; ↓: significant decrease; SCFA: short chain fatty acids; FISH: fluorescence in situ hybridization; qPCR: quantitative polymerase chain reaction.

Koutsos A, Tuohy KM, Lovegrove JA. *Nutrients*. 2015; **7**: 3959-3998

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Koutsos A, Tuohy KM, Lovegrove JA. *Nutrients*. 2015; **7**: 3959-3998

There is Bread, and There is Bread…

- Reducing white bread, but not whole-grain bread consumption within a Mediterranean-style food pattern setting is associated with lower gains in weight and abdominal fat
- It appears that the different composition between whole-grain bread and white bread varies in its effect on body weight and abdominal fat
- The term 'whole-grain bread' needs to be defined for use in epidemiological and clinical studies

Naturopathic Principle 3

- Acknowledge the NON-FOOD dietary substances
- In some cases, after processing, it is no longer considered a FOOD:
  - It becomes a chemical entity
  - It needs to be considered as having significant physiologic effects
  - As with all drugs or poisons, should be dose controlled

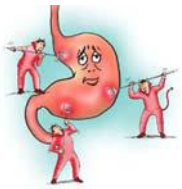
HANDLE NON-FOOD SUBSTANCES WITH CARE

Naturopathic Principle 4

Support gastrointestinal health

- The two-way obesity model that considers only the interplay between humans and their environment now to include the gastrointestinal microbiota.
- Notable perturbations in the bacterial communities in obese individuals so microbiota play a role in obesity development and propagation.
- Microbiota in the obese harvests energy more effectively
- Microbia may manipulate host gene function leading to increased adiposity
- Microbe ratios aggravation of inflammatory mechanisms
- Microbes contribute to metabolic endotoxemia
- Microbes can contribute to metabolic dysfunction

HEALTHY MICROFLORA ESSENTIAL



Graham C, Mullen A, Whelan K. *Nutr Rev.* 2015; **73**(6): 376-85

Naturopathic Principle 5

- Glycemic Index Meals – “transcends all diets”
- All body types and health conditions respond positively to a low glycemic index diet
- Ratio 2 LGI to 1 HGI meal
- Exceptions –high calorie requirements
- LOW GLYCEMIC FOODS RULE

Naturopathic Principle 5

- Effect of GI on biochemical parameters, food intake, energy metabolism, anthropometric measures, body composition in overweight subjects over 45 days.
  - Low GI meals increase fat oxidation, reduce waist circumference and HOMA-IR
  - High GI meals decrease daily dietary fiber and increase energy intake
- Low GI meal after a High GI meal:
  - Higher reduction on waist circumference and body fat, a higher increase of postprandial fat oxidation in response to the Low GI meals than after High GI meals
- High GI meals increase fasting respiratory coefficient compared to Low GI meals
- Consumption of two daily Low GI meals has a positive effect on obesity control; the consumption of High GI meals had the opposite effect

Pereira EV, Costa Jde A, Alfenas Rde C. *Arch Endocrinol Metab*. 2015; **59**(3): 245-51

Naturopathic Principle 6

Respect ORGANOLEPTIC properties of food…

<u>Prepare for digestion</u>

Naturopathic Principle 7

- Sequential eating during meal for proper food combining
- "Strata digestion" of food
- Eat smaller amounts and fewer varieties
- The smaller the amount of a particular food eaten, the less is the digestive time for that food
- The less foods mixed and the fewer the varieties used, the easier it is to digest and less tempted to overeat
- The greater the variety, the greater the tendency to overeat
- CORRECT FOOD COMBINING

Naturopathic Principle 7

- COMBINATIONS TO AVOID
  - Mixing dried sweet fruit, honey, maple syrup or bananas with nuts or seeds
  - Mixing starch foods with fresh or acid foods or fruits
  - Mixing dried sweet fruits with acid fruits
  - Eating dried sweet fruits with or after concentrated proteins
  - Eating raw, fresh or dried fruits after any cooked food
  - Avoid drinking beverages or even water during or after meals

Naturopathic Principle 8

- Family (sit down) meals instead of eating alone and on-the-run
- Social, emotional eating
- Adolescents that eat meals in a family meal are less obese that those that don't sit down family meals
- Digestion requires peace and happiness

Naturopathic Principle 9

- Regular physical movement
  - Every metabolic parameter responds to exercise
  - Oxygen essential to metabolism
  - Aim to move for 15 min every hour
  - Be conscious of time spent sitting, and move!
  - Move after meals (diabetes no more…)

Naturopathic Principle 9

- Study: Clinical Effectiveness of Weight Loss and Weight Maintenance Interventions for Men (The ROMEO Project)
  - Group interventions produced favorable weight loss results
  - Average reported participant retention rate was 78.2%, ranging from 44% to 100% retention, indicating that, once engaged, men remained committed to a weight loss intervention
  - Weight loss for men is best achieved and maintained with the **combination of a reducing diet, increased physical activity, behavior change techniques**

Robertson C, Avenell A, Stewart F, et al. *Am J Mens Health*. 2015. pii: 1557988315587550

Naturopathic Principle 9

- Study: Tour of Andalusia, a four-stage race over 647.6 km
  - The nutritional intake of the cyclists was as follows:
    - CHO, 12.8 ± 1.7 g/kg of body weight (BW; 62.3%);
    - Fat, 2.1 ± 0.2 g/kg BW (23.2%);
    - Proteins, 3.0 ± 0.3 g/kg BW (14.5%);
    - Total kcal was 5644.3 ± 593.1
  - % body fat and fat weight decreased, weight of muscle mass stayed same
  - Concentrations of urea, aspartate aminotransferase, alanine aminotransferase, creatine kinase, myoglobin, and high-density lipoproteins significantly increased

Morente-Sánchez J, Zandonai T, Mateo-March M, et al. *Scand J Med Sci* Sports. 2015; **25**(4): e423-31

Naturopathic Principle 10

- Aging changes nutrient requirements
  - Less energy expenditure, so less requirement for CHO
  - Maintenance of nerve cell function (cognitive function / reflexes, retaining bone (calcium) and muscle (protein)

Increased requirement for proteins and essential fats

Ispoglou T, White H, Preston T, et al. *Eur J Clin Nutr*. 2015. doi: 10.1038/ejcn.2015.91. [Epub ahead of print]

Naturopathic Principle 10

- The PREvención con Dieta MEDiterránea RCT of long-term nutrition intervention in subjects at high cardiovascular risk
- 15 g walnuts, 7.5 g almonds and 7.5 g hazelnuts
  - 50% reduction in incident diabetes
  - 50% reduction in incident stroke
  - 30% reduction in CVD

Ros E. *Br J Nutr*. 2015; **113** Suppl 2:S111-20

Naturopathic Principle 10

- Paleolithic nutrition improves plasma lipid concentrations of hypercholesterolemic adults to a greater extent than traditional heart-healthy diet
- Significantly lowered mean total cholesterol, LDL and TG, and increased HDL, independent of changes in body weight

Pastore RL, Brooks JT, Carbone JW. *Nutr Res.* 2015; **35**(6): 474-9

Naturopathic Principle 10

- Low calorie Mediterranean diet and advanced glycation end-products (AGEs)
  - Mean body weight, body fat, waist circumference, total cholesterol, TRI and and carboxymethyl-lysine (CML) serum levels fell significantly
  - Significant changes in CML and insulin resistance were observed in 17 women classified as compliant to caloric restriction, but not in the 27 participants who were considered adherent to the MD

Rodríguez JM, Leiva Balich L, Concha MJ, et al. *Nutr Hosp*. 2015;**31**(6): 2511-7

Naturopathic Principle 10

- Adequate protein intake is essential to retaining muscle and maintaining physical function, especially in the elderly
- L-Leucine has received attention as an essential amino acid (EAA) that enhances protein retention
- Twice-daily supplementation of EAAs containing 20% or 40% L-Leucine :
  - Improved aspects of **functional performance** and
  - At the higher level **improved lean tissue mass**

Ispoglou T, White H, Preston T, et al. *Eur J Clin Nutr*. 2015. doi: 10.1038/ejcn.2015.91. [Epub ahead of print]

Naturopathic Principle 10

- High-protein/low-carbohydrate vs standard hypocaloric diet on adipocytokine levels and cardiovascular risk factors during 9 months, role of rs6923761 gene variant of glucagon-like peptide 1 receptor:
  - Genotype GG 44.5% (wild group)
  - Genotype GA 42.2%
  - Genotype AA (13.3 %) (mutant group)
  - Lack of association of rs6923761 GLP-1 R polymorphism with weight loss
  - With both diets and in both genotype groups, body mass index, weight, fat mass, waist circumference, BP, and cholesterol, LDL, insulin levels and HOMA-R decreased

de Luis DA, Aller R, Izaola O, et al. *J Endocrinol Invest*. 2015. [Epub ahead of print]

Naturopathic Principle 11

- Gender differences in energy metabolism
- Overestimation of energy expenditure in women
- Hormonal controls affect metabolism differently
  - Oestrogen/testosterone influence LDL/HDL levels
  - Stress hormone (cortisol/DHEA) differences

Take into account metabolic differences in men and women!

Naturopathic Principle 11

- Lipid profile response is related to age and gender:
  - 96 women and 84 men overweight/obese aged 18-50yrs for 22 wks
  - 3 times/wk training for 22 weeks, 2 weeks for pre- and post evaluation
  - hypocaloric diet (25-30% less energy intake)
- Both genders had differences in HDL, LDL, triglycerides after intervention
- Men achieve a positive, greater change on lipid profile than women
- The favorable lipid profile response decreased with increasing age

Romero-Moraleda B, Peinado Lozano AB, Morencos Martínez E, et al. *Nutr Hos*p. 2015;**31**(6): 2455-64.

Naturopathic Principle 11

- Weight loss program on body composition/metabolic profile in obese postmenopausal women displaying various obesity phenotypes: a MONET group study
  - Obesity phenotypes :
  1. Lower VF and lower LBMI
  2. Lower VF and higher LBMI
  3. Higher VF and lower LBMI
  4. Higher VF and higher LBMI
- All groups had significantly improved measures of body composition after intervention
- No difference was observed among the different obesity phenotypes regarding improvements in the metabolic profile in response to weight loss

Normandin E, Doucet E, Rabasa-Lhoret R,et al. *Appl Physiol Nutr Metab*. 2015; **40**(7): 695-702.

Naturopathic Principle 12

- Adequate and correct timing of sleep
  - Time allocated to sleeping
  - Rhythm of sleep (DRS)
  - Affects energy intake
  - Affects energy expenditure

Tune into the SUN and MOON cycles

Capers PL, Fobian AD, Kaiser KA et al. Obes Rev. 2015; **16**(9): 771-82

Naturopathic Principle 12

- Study: Relationship between sleep duration and body composition, food intake or biomarkers related to food intake
  - Systematic literature review using six databases to Aug 2014
- Conclusion:
  - Sleep restriction increases food intake and total energy expenditure with inconsistent effects on integrated energy balance

Capers PL, Fobian AD, Kaiser KA et al. Obes Rev. 2015; **16**(9): 771-82

Health in the 21st Century

12 Naturopathic Principles

Traditional, supported by science.

Part 2

- From the era of modern science, understanding food compositions that are best for our genetic profile, as well of the **effect of dietary intervention on gene expression** will also be essential knowledge to 21st century nutritional medicine.

Nutrigenomics

Science supports naturopathic principles.

Principles of Nutrigenomics

- Overview of different ways nutrition and genome are inter-connected
- Influence of nutrigenomics on clinical practice

Does current nutrigenomic research support naturopathic clinical practice?

Principles of Nutrigenomics

Table 1. Nutrition is complicated due to many variables.

- We eat ~1.5 kg food & drink ~2 L liquid/day;
- About 40 essential nutrients are known;
- Thousands of known compounds in foods without known biological functions;
- Thousands of unknown compounds in foods without known biological functions;
- About 10^{13} cells in the body & about 10^{14} bacteria in the GI tractus;
- Mostly unknown and complicated interplay between diet and the microbiome;
- Many organs & some hundreds of cell types are found in the body;
- About 25,000 genes in human cells;
- Human genome includes 3 billion base pairs;
- Some millions single nucleotide polymorphisms (SNPs);
- A large epigenetic variation between individuals due to environmental factors;
- About 100,000 transcripts (mRNA);
- About 100,000 proteins;
- About 1000 lipids & thousands of water-soluble metabolites.

Norheim F, Gjelstad IM, Hjorth M, et al. *Nutrients* 2012; **4**(12): 1898-1944

Principles of Nutrigenomics

- The Human Variome Project (HVP)
- http://www.humanvariomeproject.org
- International effort to identify genes, their mutations, and their variants associated with phenotypic variability and indications of human disease or phenotype
- Any two humans will differ by 3-5 million bases and re-sequencing of individual genomes has now identified 18 million single nucleotide polymorphisms

Principles of Nutrigenomics

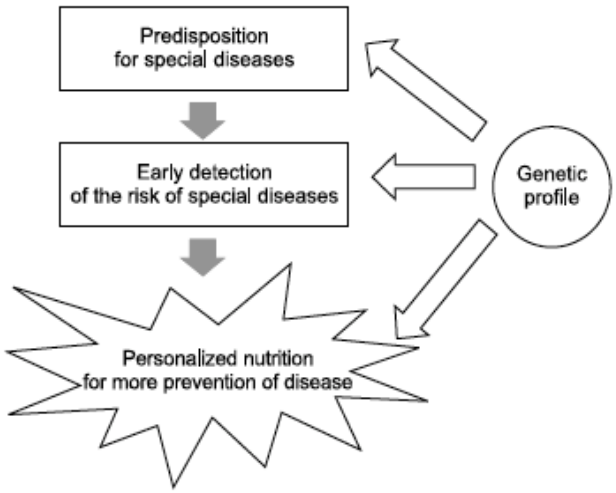
Nutrigenomics:

- Study of the unidirectional interactions between genes and diet
- Understanding of how genetic variation and epigenetic events alter requirements for, and responses to, nutrients

Principles of Nutrigenomics

- Science of nutrigenetics and nutrigenomics
- Nutrition may exert its impact on health outcomes by:
  - Directly affecting expression of genes in critical metabolic pathways; and/or
  - Indirectly affecting the incidence of genetic mutation at the base sequence or chromosomal level, which in turn causes alterations in gene expression
- The health effects of nutrients and nutriomes (nutrient combinations) depend on inherited genetic variants that alter the uptake and metabolism of nutrients and/or the molecular interaction of enzymes with their nutrient cofactor, and hence the activity of biochemical reaction

Principles of Nutrigenomics



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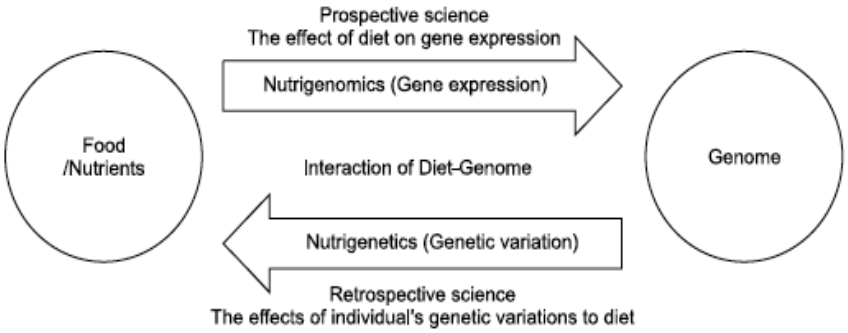
Principles of Nutrigenomics



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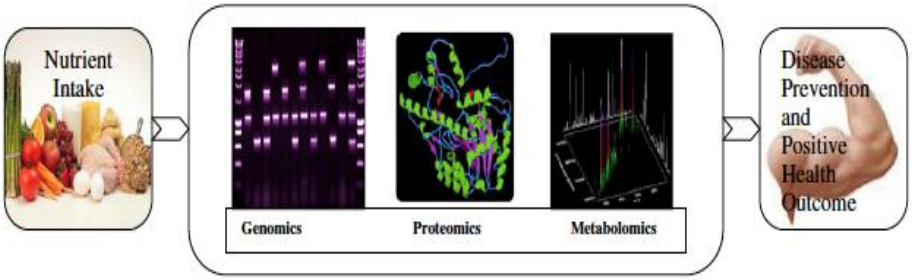
Principles of Nutrigenomics



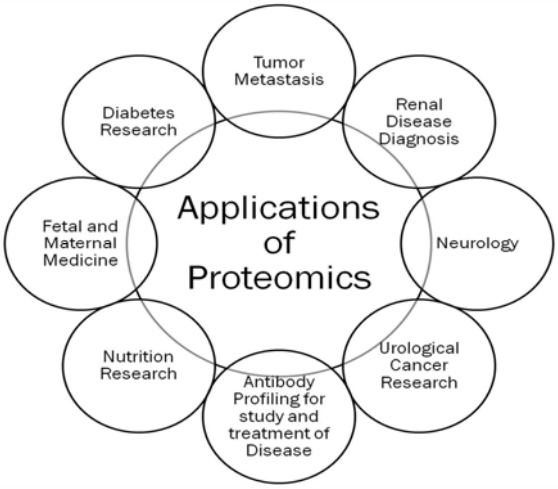
Neeha VS, Kinth P. *J Food Sci Technol*. 2013; **50**(3): 415–428

Principles of Nutrigenomics

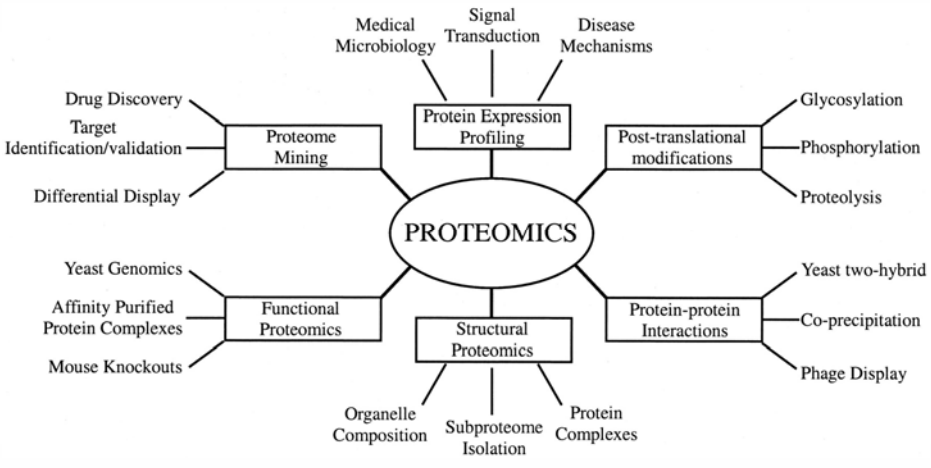
- These proteins act in the cell, tissue, or organ in its normal state, but in different physiological or pathological situations, they may change their expression level, or even their activity
- **Proteomics**
  - Proteomics is the science that studies the complete set of proteins involved in the biological processes of a certain species

Sales NM, Pelegrini PB, Goersch MC. *J Nutr Metab*. 2014; Article ID 202759

Principles of Nutrigenomics



Principles of Nutrigenomics



Sales NM, Pelegrini PB, Goersch MC. *J Nutr Metab*. 2014; Article ID 202759

Principles of Nutrigenomics

- **Metabolomics**
  - Integrated study of the many small molecules produced by metabolism
  - Methods for profiling almost all of the products of metabolism in a single sample of blood or urine are now being developed

Sales NM, Pelegrini PB, Goersch MC. *J Nutr Metab*. 2014; Article ID 202759

Principles of Nutrigenomics

- Primary metabolites are directly involved with synthesis and degradation of macromolecules
- They act as substrates, inhibitors/activators of an enzyme, molecular precursors, wastes of synthesis, or degradation of macromolecules
- Metabolomics allows understanding of metabolic arrangements and instabilities that cause or suffer interference from the diet

Sales NM, Pelegrini PB, Goersch MC. *J Nutr Metab*. 2014; Article ID 202759

Principles of Nutrigenomics

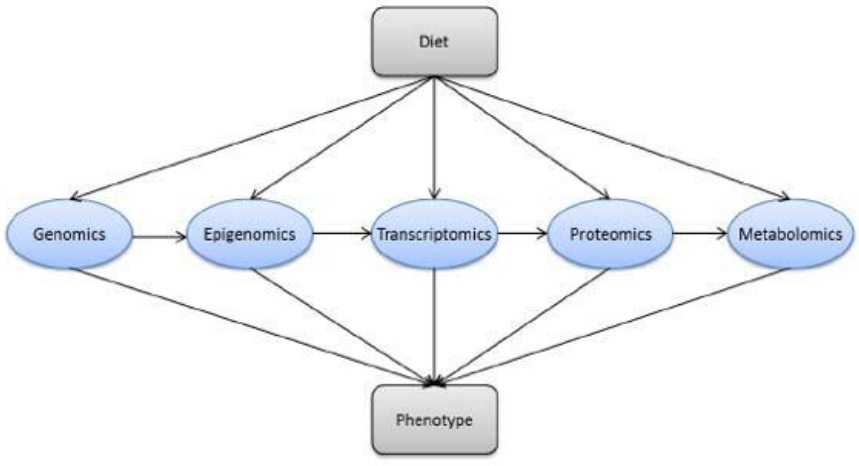
Epigenetics

- Processes that regulate how and when certain genes are turned on and off

Epigenomics

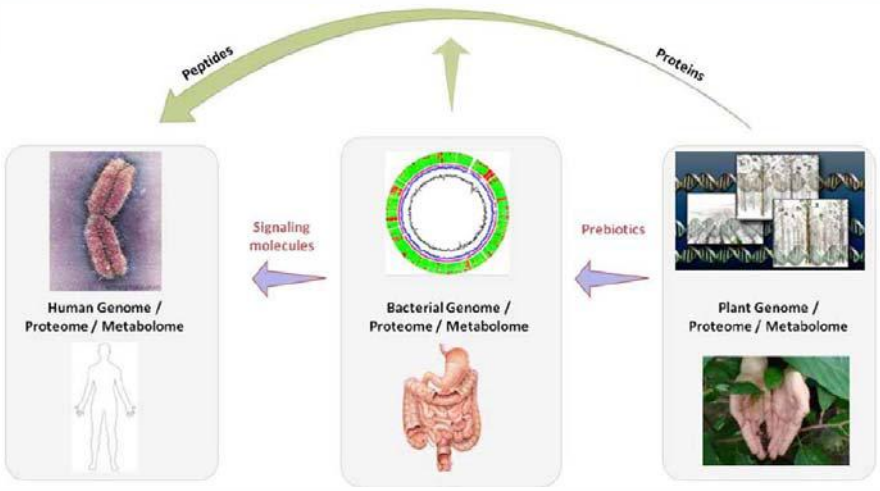
- Analysis of epigenetic changes in a cell or entire organism

Principles of Nutrigenomics



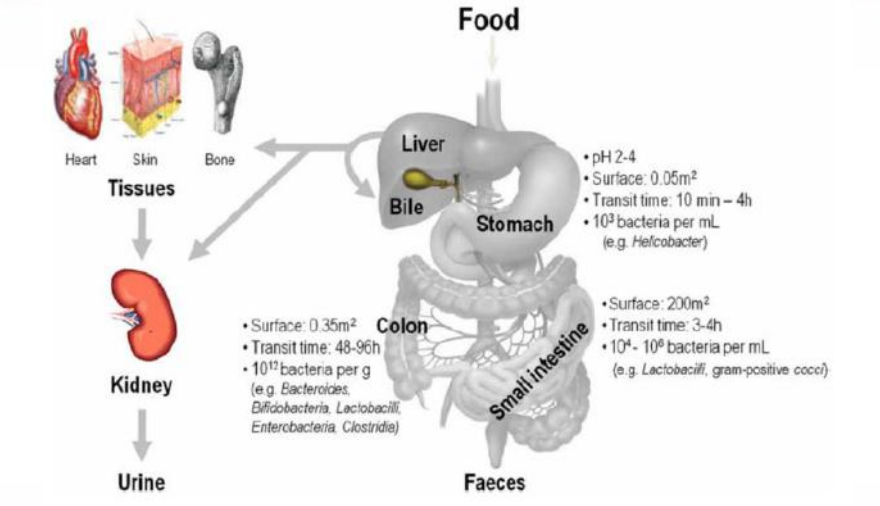
Neeha VS, Kinth P. *J Food Sci Technol.* 2013; **50**(3): 415–428

Principles of Nutrigenomics



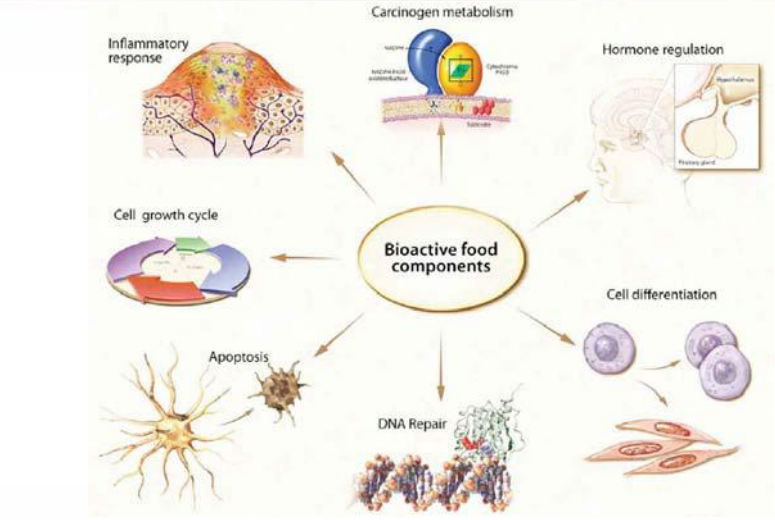
Kussmann M, Van Bladeren PJ. *Front Genet*. 2011; **2**: 21.

Principles of Nutrigenomics



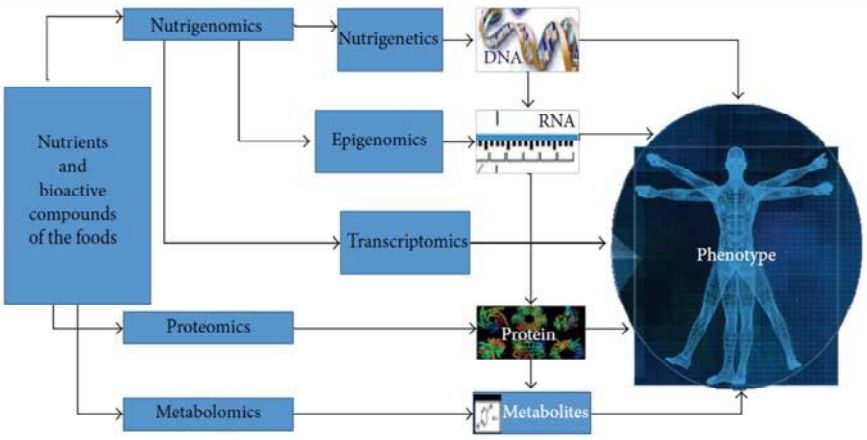
Kussmann M, Van Bladeren PJ. *Front Genet*. 2011; **2**: 21.

Principles of Nutrigenomics



Trujillo E, Davis C, Milner J. *J Am Diet Assoc*. 2006; **106**(3): 403-413

Principles of Nutrigenomics



Sales NM, Pelegrini PB, Goersch MC. *J Nutr Metab*. 2014; Article ID 202759

Principles of Nutrigenomics

- Nutrigenomics and clinical research:
  - Nutrition studies are fraught with problems
  - Assumptions for dietary requirements, energy needs based on age & gender
  - Assumptions do not plan for the array of suboptimal nutritional deficiencies or excesses that exist and may impact on outcomes
  - This leads to too much 'noise' and favors a null hypothesis in trials

Principles of Nutrigenomics

- Nutrigenomics will provide understanding of metabolism to establish the correct research programs to answer the clinical trial questions
- Identify responders and non-responders on the basis of their genetic profile
- Target groups can be identified for intervention studies
- Scientists – depth of understanding of metabolism means we play a role establishing the correct research programs to answer the clinical trial questions

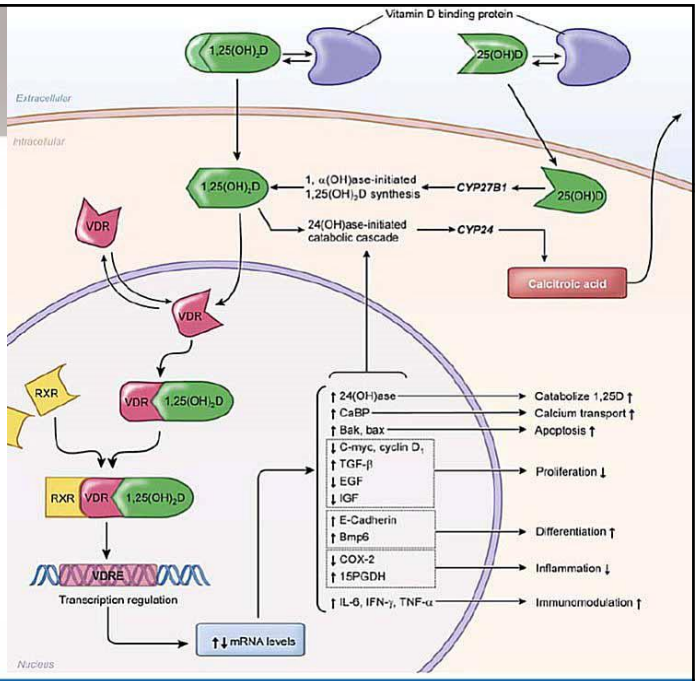
Principles of Nutrigenomics

- Nutrigenomics and clinical practice:
  - Better research methods ensure a new foundation for clinical nutrition
  - Enables subsequently individualized dietary recommendations
  - Provides understanding to why specific treatments are effective or not

Principles of Nutrigenomics

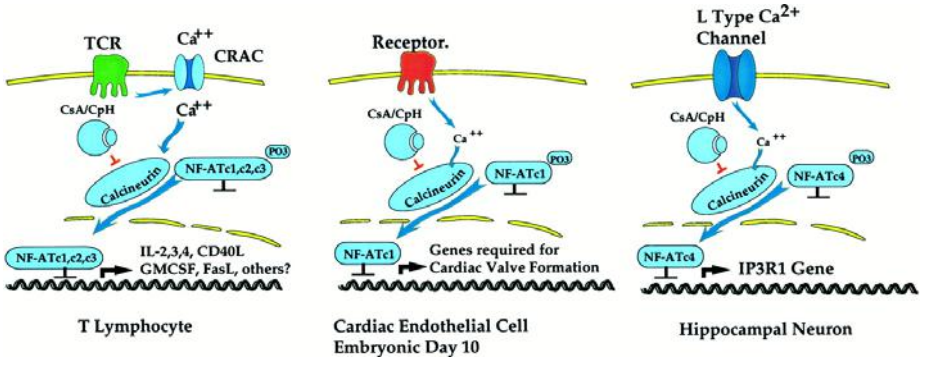
- Effect through transcription:
  - Direct interactions: nutrients, sometimes after interacting with a receptor, behave as transcription factors that can bind to DNA and acutely induce gene expression
  - Nutrients and dietary components that can alter genetic and epigenetic events, and thereby influence health include:
  - Vitamin D, Calcium, vitamin A, Fatty acids, Zinc
  - Selenium, folate, vitamin C and vitamin E
  - Bioactives - Linolenic acid, n-3-fatty acids,
  - Phytochemicals - flavonoids, indoles, isothiocyanates, carotenoids
  - Fungochemicals - B-glucans
  - Bacteriochemicals - butyrate, probiotic fermentation products

Vitamin D and the Vitamin D Receptor



Davis & Milner *J Nutrigenet Nutrigenomics* 2011; **4**: 1–11

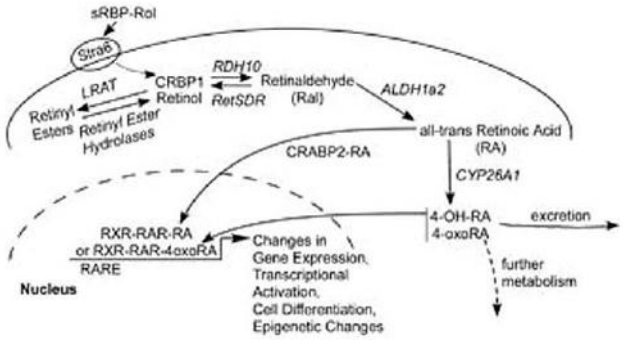
Nutrigenomics: Calcium and Calcineurin (PP2B)



Crabtree GR, *J Biol Chem.* 2001; **276**(4): 2313–2316

Nutrigenomics: Vitamin A and Retinoic Acid Receptor Proteins

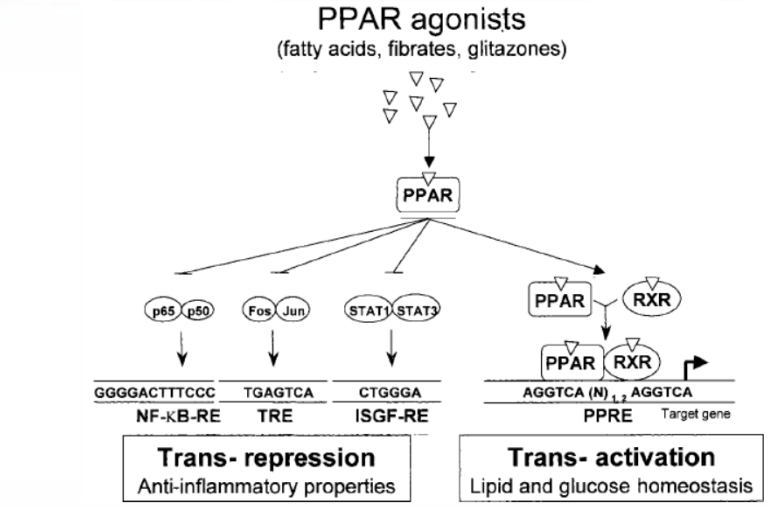
These complexes activate or repress transcription



Uptake and intracellular metabolism of retinol (modified from Mongan and Gudas, [2007]).

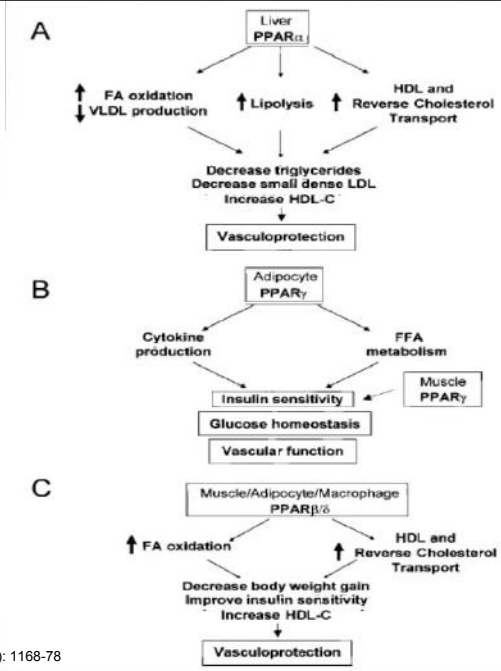
Gudas LJ, Wagner JA. *J Cell Physiol*. 2011; **226**(2): 322–330

Nutrigenomics: Fatty Acids and PPARα



Marx N, Duez H, Fruchart JC, et al. *Circ Res.* 2004; **94**(9): 1168-78

Nutrigenomics: Fatty Acids and PPARα



Marx N, Duez H, Fruchart JC, et al. *Circ Res.* 2004; **94**(9): 1168-78

Nutrigenomics: Fatty Acids

Gene Name	Function / Comments
FABP4	fatty acid binding protein 4, adipocyte, adipocyte fatty acid binding protein P2 (aP2); regulator of toxic lipid-induced ER stress; is secreted from adipocytes and influences hepatic glucose homeostasis
UCP1	uncoupling protein 1; major determinant of brown adipocytes (BAT), involved in adaptive thermogenesis by acting as an uncoupler of mitochondrial oxidative phosphorylation
PLIN1	perilipin 1; hormonally-regulated protein on the surface of fat droplets; phosphorylated by PKA and then directs PKA-activated hormone-sensitive lipase (HSL) to diacylglycerides in the droplets
Adipocytokines	
Gene Name	**Function / Comments**
ADPN	adiponectin; also known as adipoQ for adipocyte, C1q and collagen-domain containing protein
Lipogenesis	
Gene Name	**Function / Comments**
ACACA	acetyl-CoA carboxylase 1 (ACC1); acetyl-CoA carboxylase-α (alpha); major rate-limiting enzyme of fatty acid synthesis
ELOVL4	elongation of very long-chain fatty acids-like 4; required for the synthesis of very long chain saturated fatty acids; also required for very long chain polyunsaturated fatty acid synthesis that are unique to retina, sperm, and brain
LXRA	liver X receptor α (alpha)
ME1	malic enzyme 1; involved in the pathway by which acetyl-CoA is transported out of the mitochondria as citrate, converts cytosolic malate to pyruvate while also generating NADPH
SCD1	stearoyl-CoA desaturase 1, (delta) Δ^9-desaturase

Marx N, Duez H, Fruchart JC, et al. *Circ Res.* 2004; **94**(9): 1168-78

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Marx N, Duez H, Fruchart JC, et al. *Circ Res.* 2004; **94**(9): 1168-78

Nutrigenomics: Fatty Acids

Lipid Binding & Transport / Lipoproteins	
Gene Name	**Function / Comments**
APOA2	apolipoprotein A-II; primary apoprotein of HDL; activates hepatic lipase
APOE	apolipoprotein E; important for recognition of lipoprotein particles by the LDL receptor; essential for metabolism of triglyceride-rich lipoproteins
CD36	leukocyte differentiation antigen 36; also known as fatty acid translocase, FAT; is a lipoprotein scavenger receptor as well as a membrane fatty acid transporter
LDLR	LDL receptor
LIPC	hepatic lipase, hepatic triglyceride lipase (HTGL); synthesized by heptocytes and bound to hepatic endothelial surfaces via heparin sulfate proteoglycans (HSPGs); catalyzes hydrolysis of fatty acids at the *sn*1 position of phospholipids and of mono-, di-, and triacylglycerides associated with a variety of lipoproteins
LRP1	LDL receptor-related protein 1
LPL	lipoprotein lipase, bound to vascular endothelial cell surfaces; predominantly expressed in cardiac and skeletal muscle and adipose tissue; catalyzes hydrolysis of fatty acids at the *sn*1 and *sn*2 position of phospholipids and of mono-, di-, and triacylglycerides associated with a variety of lipoproteins
OLR1	oxidized LDL (oxLDL) receptor; also identified as the endothelial oxLDL receptor: LOX-1
FATP1	fatty acid transport protein 1; very long-chain acyl-CoA synthetase family member 4 (ACSVL4); solute carrier family 27, member 2 (SLC27A1); integral membrane protein involved in uptake of long-chain and very long-chain fatty acids along simultaneous with CoA activation
FATP2	fatty acid transport protein 2; very long-chain acyl-CoA synthetase (VLACS); solute carrier family 27, member 2 (SLC27A2); integral membrane protein involved in uptake of long-chain and very long-chain fatty acids along simultaneous with CoA activation

Marx N, Duez H, Fruchart JC, et al. *Circ Res.* 2004; **94**(9): 1168-78

Nutrigenomics: Fatty Acids

Glucose Homeostasis	
Gene Name	**Function / Comments**
G6PC	glucose-6-phosphatase, catalytic
GPD1	glycerol-3-phosphate dehydrogenase 1, cytosolic; enzyme of glycerol-phosphate shuttle used to transfer cytosolic NADH into the mitochondria; also involved in the synthesis of triglycerides in adipose tissue
GCK	glucokinase
PEPCK	phosphoenolpyuvate carboxykinase
PDK4	pyruvate dehydrogenase kinase 4; expressed in all tissue but with highest levels cardiac and skeletal muscles

Marx N, Duez H, Fruchart JC, et al. *Circ Res.* 2004; **94**(9): 1168-78

Nutrigenomics: Fatty Acids

Lipid Binding & Transport / Lipoproteins	
Gene Name	**Function / Comments**
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LRP1	LDL receptor-related protein 1
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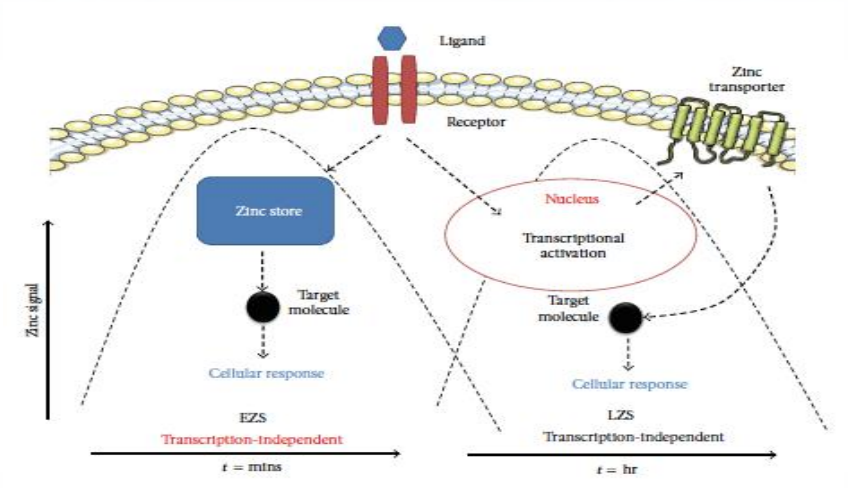
Marx N, Duez H, Fruchart JC, et al. *Circ Res.* 2004; **94**(9): 1168-78

Nutrigenomics: Zinc and Metal Responsive Transition Factor-1 (MRTF-1)



Myers et al. *Journal of Nutrition and Metabolism* 2012; Article ID 173712

Principles of Nutrigenomics

2. DNA and epigenetic alterations
   - Nutrients can alter the structure of DNA (or of histone proteins in chromatin) so that gene expression is chronically altered
     - They are mediated by methylation of DNA or by methylation, acetylation, or biotinylation of histones
     - Result in changes in gene expression that can last throughout a person's life and can even persist across generations
     - Because a significant portion of human genes are regulated by methylation, many more examples should accrue of diet-related changes in gene methylation

Principles of Nutrigenomics

- Longer survival and a 75% lower risk of DM in whose paternal grandfathers experienced food scarcity during the slow growth period just before puberty than in those whose paternal grandfathers did not
- These effects of nutrition must occur via epigenetic imprinting of paternal genes (when the first viable pools of spermatocytes emerge and when reprogramming of DNA methylation imprinting begins)

Principles of Nutrigenomics

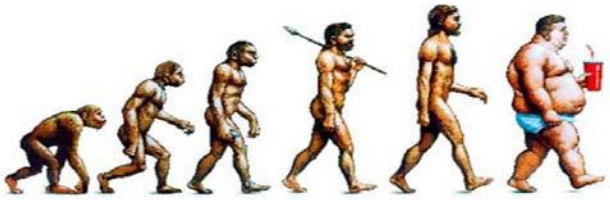
- Dutch Famine Cohort - shown that serious nutritional deprivation during pregnancy caused an increased risk of metabolic disorders in offspring several decades later
- This period of maternal starvation was very significant and caused marked differences in disease outcomes; the first trimester of pregnancy was particularly vulnerable to disease outcome in adulthood
- There is a critical developmental time window where dietary pattern can induce epigenetic changes
- These epigenetic changes are passed to offspring

de Rooij SR, Painter RC, Roseboom TJ, et al. *Diabetologia*. 2006; **49**(4): 637-43

Principles of Nutrigenomics

Evolution of the Human Race Continues…

Homo obesus



Homo sapiens > Homo obesus

Temelkova-Kurktschiev T, Stefanov T. *Exp Clin Endocrinol Diabetes* 2012; **120**: 1–6

Principles of Nutrigenomics

3. Genetic variations (SNP's)
   - Common genetic variations [single-nucleotide polymorphisms (SNPs) can alter the expression or functionality of genes and influence individual nutrient requirements
   - A database of functionally important SNPs would make it possible for a nutrition specialist— one who has an understanding of the effects of such changes on metabolism and nutrient requirements—to use SNP profiling to make practical recommendations as part of clinical practice (eg. a recommendation of higher dietary choline intake during pregnancy in women with SNPs that alter PEMT expression)

Zeisel SH. *Am J Clin Nutr.* 2007; **86**: 542-548

Principles of Nutrigenomics

- Gene polymorphisms
  - Small sequence difference in a gene
  - They affect how well a protein works and how the protein interacts with other proteins or substrates
- SNP's
  - A haplotype is the set of SNP alleles along a region of a chromosome
  - Haplotype analysis can be used to identify groups of SNPs linked together, and useful in understanding the distribution of risk alleles in populations and for tailoring prevention strategies to those at increased risk
  - Particular to metabolism: Many SNPs in the human growth hormone secretagogue receptor (known as the GHSR or ghrelin receptor) have an important role in the regulation of food intake and energy homeostasis

Zeisel SH. *Am J Clin Nutr.* 2007; **86**: 542-548

Principles of Nutrigenomics

- Humans share the same genes, but with many individual variations in the codon sequences
- In total, 10 million SNPs exist that occur in 1% of the population
- Some common SNPs occur in 5-50% of the population
- Most humans are heterozygous for 50,000 SNPs across their genes
- Some fraction of these SNPs results either in alteration of gene expression or in changes in the gene product such that protein structure and function are altered
- Mastering the ways in which millions of SNPs may influence nutrient requirements is daunting
- A number of relatively common SNPs are known to influence nutrient requirements

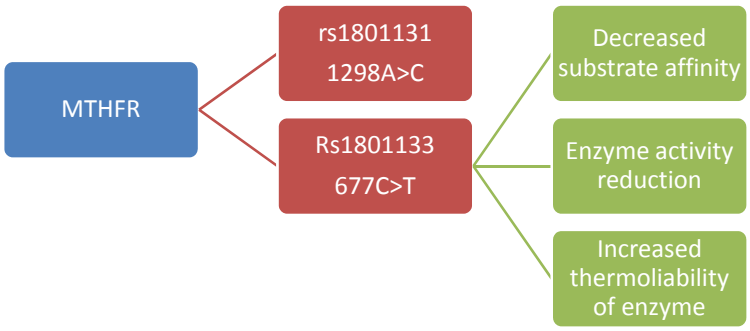
Joslin AC, Green R, German JB, et al. *Genes Nutr*. 2014; **9**(5): 419

Principles of Nutrigenomics

- The enzyme 5,10-methylenetetrahydrofolate reductase (MTHFR) - folate metabolism
- The MTHFR gene has a common SNP (C677T allele) that results in reduced enzymatic activity, and homozygous persons have elevated plasma homocysteine concentrations unless they ingest high amounts of folate
- This SNP occurs in 15–30% of the population

Joslin AC, Green R, German JB, et al. *Genes Nutr*. 2014; **9**(5): 419

Principles of Nutrigenomics



Adapted from Joslin AC, Green R, German JB, et al. *Genes Nutr*. 2014; **9**(5): 419

Principles of Nutrigenomics

- Choline, genetics and endocrine and liver function
  - Some people develop fatty liver and liver and muscle damage, whereas others did not. The PEMT gene encodes for a protein responsible for endogenous formation of choline in the liver and it is induced by estrogen
  - Premenopausal women who were carriers of a very common SNP (MTHFD1-G1958A) were 15 times as likely as noncarriers to develop signs of choline deficiency on a low-choline diet
  - A recommendation of higher dietary choline intake during pregnancy in women with SNPs that alter PEMT expression

Zeisel SH. *Am J Clin Nutr.* 2007; **86**: 542-548

Principles of Nutrigenomics

- AMP-activated protein kinase (AMPK):
  - important key elements in energy control, appetite regulation, myogenesis, adipocyte differentiation, and cellular stress management
  - Critical regulator of cellular metabolism, important in diabetes, cancer, and vascular disease
- During ischemia-reperfusion, AMPK activation modulates glucose and fatty acid metabolism, mitochondrial function, ER stress, autophagy, and apoptosis
- AMPK prevents myocardial necrosis and contractile dysfunction during ischemia-reperfusion and represents a cardioprotective strategy for treatment of myocardial infarction

Qi D, Young LH. *Trends Endocrinol Metab.* 2015; **26**(8): 422-9

Clinical Nutrigenomics

Nutritional therapy re-balances the expression of genes and metabolites

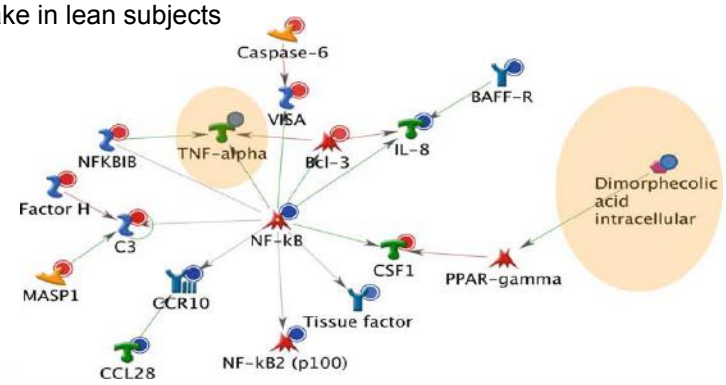
Clinical Nutrigenomics

- Fifteen lean and 17 obese men on 50g or 200g vegetables for 4 weeks, followed by 4 weeks of energy restriction (60% of normal energy intake)
- In both groups, adipose tissue gene expression changes in response to vegetable intake, including the network of inflammation genes
- In obese subjects, high vegetable intake also resulted in changes related to energy metabolism, adhesion and inflammation. **In total, 323 genes were affected in obese subjects and 532 genes in lean subjects**
- By inclusion of sensitive omics technologies and comparing the changes induced by high vegetable intake with changes induced by energy restriction, it has been shown that part of vegetables' health benefits are mediated by changes in energy metabolism, inflammatory processes and oxidative stress

Pasman WJ, van Erk MJ, Klöpping WA, et al. *Genes Nutr*. 2013; **8**(5): 507-21

Clinical Nutrigenomics

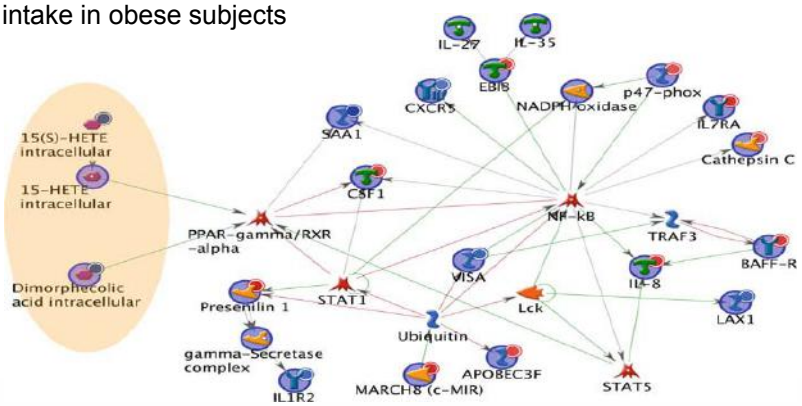
- Network showing biological links between genes involved in inflammation and plasma markers that respond to high vegetable intake in lean subjects



Pasman WJ, van Erk MJ, Klöpping WA, et al. *Genes Nutr*. 2013; **8**(5): 507-21

Clinical Nutrigenomics

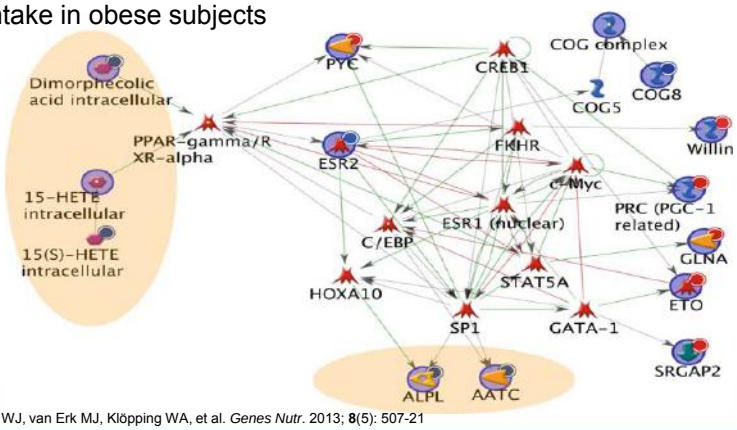
- Network showing biological links between genes involved in inflammation and plasma markers that respond to high vegetable intake in obese subjects



Pasman WJ, van Erk MJ, Klöpping WA, et al. *Genes Nutr*. 2013; **8**(5): 507-21

Clinical Nutrigenomics

- Network showing biological links between genes involved in energy metabolism and plasma markers that respond to high vegetable intake in obese subjects



Pasman WJ, van Erk MJ, Klöpping WA, et al. *Genes Nutr*. 2013; **8**(5): 507-21

Clinical Nutrigenomics

- Study:
  - For 30 participants RNA was extracted from peripheral blood mononuclear cells (PBMCs) and expression levels of 47,231 mRNA transcripts were assessed
  - The Prudent dietary pattern - high intakes of vegetables, fruits, whole grain products and low intakes of refined grain products
  - Western dietary pattern - high intakes of refined grain products, desserts, sweets and processed meats
  - When individuals with high scores for the Prudent dietary pattern where compared to individuals with low scores, 2,083 transcripts were differentially expressed in men, 1,136 transcripts in women and 59 transcripts were overlapping in men and women

Bouchard-Mercier A, Paradis AM, Rudkowska I, et al. *Nutr J.* 2013; **12**: 24.

Clinical Nutrigenomics

- For the Western dietary pattern, 1,021 transcripts were differentially expressed in men with high versus low scores, 1,163 transcripts in women and 23 transcripts were overlapping in men and women
- The genes differentially expressed for both patterns were present in networks related to the immune and/or inflammatory response, cancer and cardiovascular diseases
- Conclusion:
  - Gene expression profiles were different according to dietary patterns, which probably modulate the risk of chronic diseases

Bouchard-Mercier A, Paradis AM, Rudkowska I, et al. *Nutr J.* 2013; **12**: 24

Table 1 Factor loadings for Prudent and Western dietary patterns (n = 210)

Food groups (servings/day)	Factor 1* Prudent	Factor 2* Western
Vegetables	**0.71**	0.03
Fruits	**0.60**	-0.01
Whole grain products	**0.53**	0.21
Non-hydrogenated fat	**0.46**	0.02
Refined grain products	**-0.45**	**0.39**
Desserts	-0.01	**0.80**
Sweets	0.09	**0.77**
Beer	0.01	-0.03
Coffee	0.06	0.15
Poultry	-0.004	-0.06
Red meat	-0.11	0.11
Potatoes other than French fries	0.09	0.16
Processed meat	-0.10	**0.33**
Legumes	0.15	0.13
Tea	0.08	-0.02
High-fat dairy products	0.13	0.13
Low-fat dairy products	0.27	0.07
Eggs	0.27	-0.05
Cream soup	-0.11	0.12
Pizza	-0.23	-0.03
Fish and other sea food	0.28	-0.03
Fruit juices	-0.14	0.02
Nuts	0.26	0.06
Vegetable juices	0.12	0.05
Condiments	0.18	0.06
Snacks	-0.11	0.18
Saturated fat (butter and lard)	0.04	0.06
Variance explained (%)	12.96	10.62

*Exploratory factor analysis using the FACTOR procedure.
Factor loading ≥ 0.30 or ≤ -0.30 are marked in bold.

Bouchard-Mercier A, Paradis AM, Rudkowska I, et al. *Nutr J.* 2013; **12**: 24

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Clinical Nutrigenomics

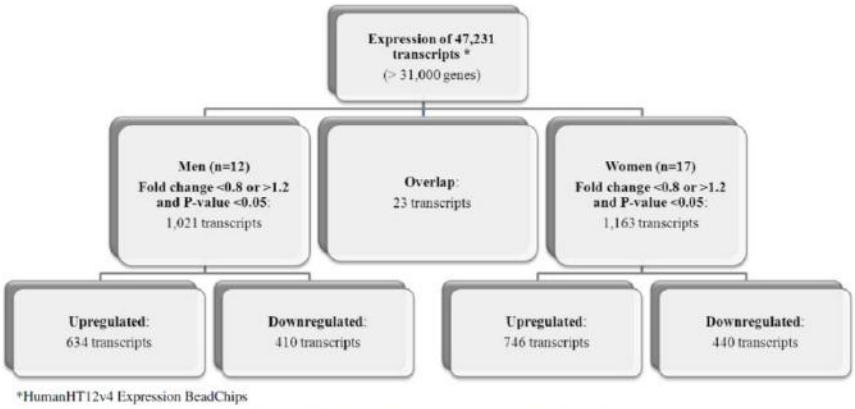
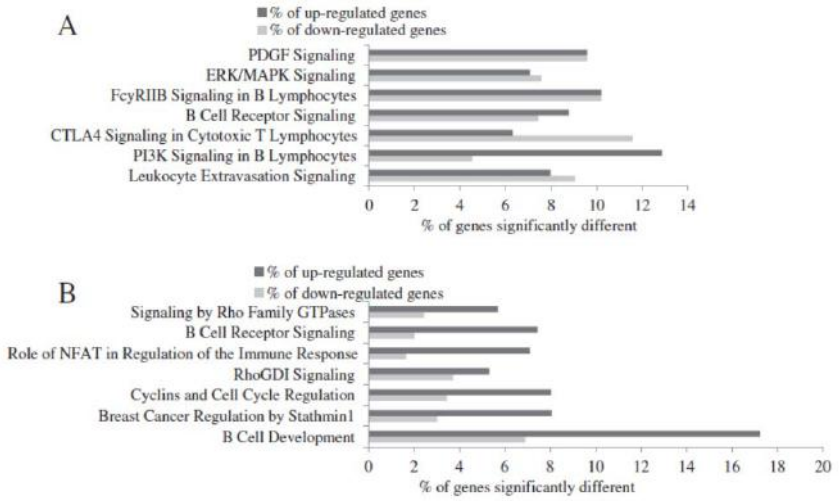


Figure 2 Flowchart illustrating the significantly different transcripts according to scores for the Western dietary pattern.

Bouchard-Mercier A, Paradis AM, Rudkowska I, et al. *Nutr J.* 2013; **12**: 24

Clinical Nutrigenomics



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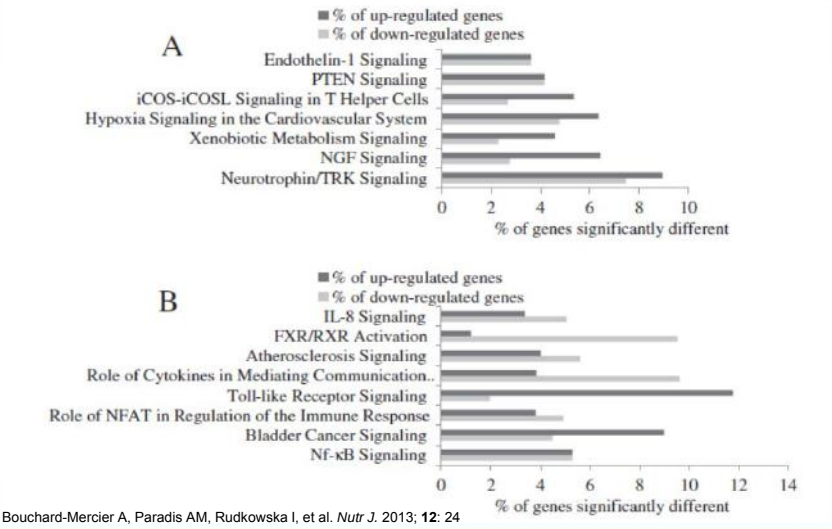
Clinical Nutrigenomics



Bouchard-Mercier A, Paradis AM, Rudkowska I, et al. *Nutr J.* 2013; **12**: 24

Clinical Nutrigenomics

- More than 40 independent T1DM-associated tagging SNPs
- Twin studies:
  - Di-zygotic twins, T1DM the concordance rate is 10%
  - Mono-zygotic twins, T1DM the concordance rate is 50%
- Dietary and environmental factors influence T1DM incidence and development:
  - Use of breast milk vs. infant formula
  - Highly hydrolyzed infant formula vs. conventional infant formula
  - Early/late exposure to gluten and casein (milk protein)
  - Nutrients can modify, alone or through changes in the gut microbiota, the expression of genes involved in the immune response
- For T1DM, nutrients may promote autoimmune responses in individuals predisposed to this condition

Berná G, Oliveras-López MJ, Jurado-Ruíz E, et al. *Nutrients.* 2014; **6**(11): 5338-69

Clinical Nutrigenomics

- Most Relevant T2DM Susceptibility Genes:
  - Variants in peroxisome proliferator-activated receptor gamma (PPARγ), TCF7L2 and fat mass and obesity-associated protein (FTO) genes
- Diets with a low glycemic load reduce the risk of T2DM conferred by TCF7L2
- TCF7L2 risk variant carriers may reduce their susceptibility to T2DM through dietary modifications

Fisher E, Boeing H, Fritsche Aet al. *Br J Nutr*. 2009; **101**(4): 478-81

Clinical Nutrigenomics

- T2DM and the FTO gene (FTO-rs9939609 variant):
  - FTO has been consistently associated with obesity risk
  - When adherence to the Mediterranean diet was low, carriers of the variant alleles showed a higher T2DM risk.
  - In contrast, when adherence to the Mediterranean diet was high, these associations disappeared
  - Patients with T2DM, carriers of the AA genotype of FTO rs9939609, showed increased fat and decreased fiber consumption, independent of BMI

Ortega-Azorín C, Sorlí JV, Asensio EM, et al. *Cardiovasc Diabetol*. 2012; **11**: 137-146
Steemburgo T, Azevedo MJ, Gross JL, et al. J *Nutrigenet Nutrigenomics.* 2013; **6**: 97-106

Clinical Nutrigenomics

- Importance of genotype by macronutrient interactions for T2DM:
  - The genome-environment contribution of 14 dietary factors to the total phenotypic variance of 4 T2DM-related traits (fasting glucose, fasting insulin, HOMA-IR and HOMA of β cell function)
  - 25.1% and 24.2% of heritability of fasting insulin and HOMA-IR explained by the genome-environment interaction of CHO intake with the whole genome
  - The heritability explained by the genome alone for fasting insulin and HOMA-IR were 20.2% and 20.9%, respectively
  - 39.0% of the heritability of HOMA of β cell function explained by the genome environment interaction of n-6 PUFA with the genome, while the heritability explained by the main effect of the genome without these interactions was 18.7% for HOMA of β cell function

Zheng JS, Arnett DK, Lee YC, et al. *PLoS One*. 2013; **8**(10): e77442.

Clinical Nutrigenomics

Clinical Application:

- T1DM / autoimmune disorders
  - Epigenetic interactions most important
  - Interventions in fetal development / childhood essential
- T2DM / non-immune related disorders
  - Direct interactions and genetic variations most important
  - Dietary changes have acute (immediate) effects
  - Consider epigenetic interactions for patient wanting to reproduce themselves

Clinical Nutrigenomics

- A number of chromosome regions have been described as associated with Inflammatory bowel disease (IBD):
  - One IBD1 site is NOD2/CARD15
- Generally, the affected genes fall into one of three classes:
  - Those affecting immune response (support with glutamine, zinc, selenium)
  - Those affecting bacterial recognition
  - Those affecting transport
- MDR1 gene encodes for P-glycoprotein (Pgp) which can be activated by herbals such as curcumin, ginsenosides, piperine, some catechins from green tea, and silymarin, while other catcehins and St. John's Wort inhibit expression

Ferguson LR, Peterman I, Hübner C, et al. *Genes Nutr*. 2007; **2**(1): 71-3.

Clinical Nutrigenomics

- A turmeric extract beneficially affected the variants of SLC22A4 and IL-10 associated with IBD, by reducing inappropriate epithelial cell transport (SLC22A4, 503F) and increasing anti-inflammatory cytokine gene promoter activity (IL-10, −1082A)
- The effect of turmeric on the IL-10 variant was strongly associated with the curcumin content of the extract and its fractions

Nutrients 2014, 6, 4178-4190; doi:10.3390/nu6104178

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nutrients

ISSN 2072-6643

www.mdpi.com/journal/nutrients

Article

The Effect of Turmeric (*Curcuma longa*) Extract on the Functionality of the Solute Carrier Protein 22 A4 (SLC22A4) and Interleukin-10 (IL-10) Variants Associated with Inflammatory Bowel Disease

Mark J. McCann [1,2,3,*], Sarah Johnston [1,2], Kerri Reilly [1,2], Xuejing Men [1,2,3,4], Elaine J. Burgess [2,5], Nigel B. Perry [2,5] and Nicole C. Roy [1,2,3,4]

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Received: 19 July 2014; in revised form: 9 September 2014 / Accepted: 29 September 2014 / Published: 13 October 2014

McCann MJ, Johnston S, Reilly K, et al. *Nutrients*. 2014; **6**(10): 4178-90.

Nutrigenomics

Science supports naturopathic principles.

Nutri-Ethics

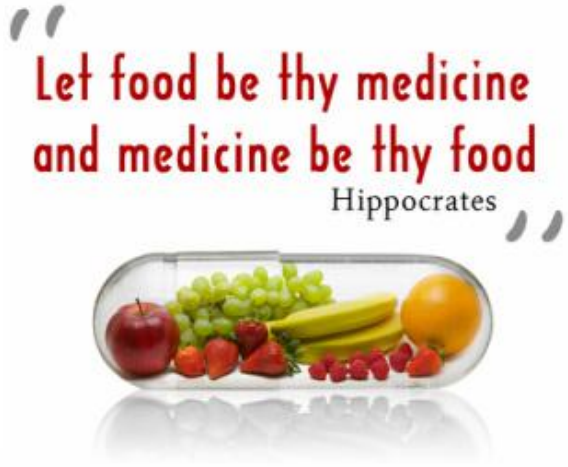
Nutri-ethics:
Study of ethics issues embedded in nutrigenomics and various intersections of food science

Form of personalized doping?
Specifically tailored to the genomics and proteomics makeup of the athlete (nutriproteomics)

Why We Eat What We Eat?

- Humans have a hedonistic mind-set, inclined to pursue happiness
- The food intake pattern of an individual is a form of behavior, frequency of eating and size of portions. This behavioral pattern, together with nutrient content and energy density of the diet, determines the amount of energy that is ingested, which promote eating even without energetic requirements for food

Lenoir M, Serre F, Cantin L, et al. PLoS One. 2007; **2**(8): e698
Volkow ND, Wang GJ, Baler RD. *Trend Cogn. Sci*. 2011; **15**: 37-46
Keijer J, Hoevenaars FP, Nieuwenhuizen A, et al. *Nutrients*. 2014; **6**: 4531-4551

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INTEGRIA HEALTHCARE SYMPOSIUM 2015

Clinical Pearls + Practitioner Perspectives

An Experience-Based Approach to Patient Care

24+25 OCTOBER
MELBOURNE

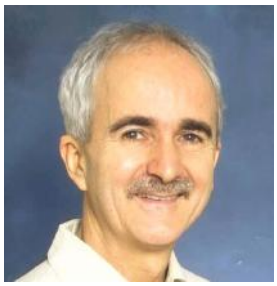
Clinical Pearl:
('klɪnɪkəl pûrl)

1. A straightforward and meaningful piece of clinical advice
2. Clinically relevant information based on experience

Session 7

Natural Solutions for Neurological Health and Cognition: New Answers to Old Questions

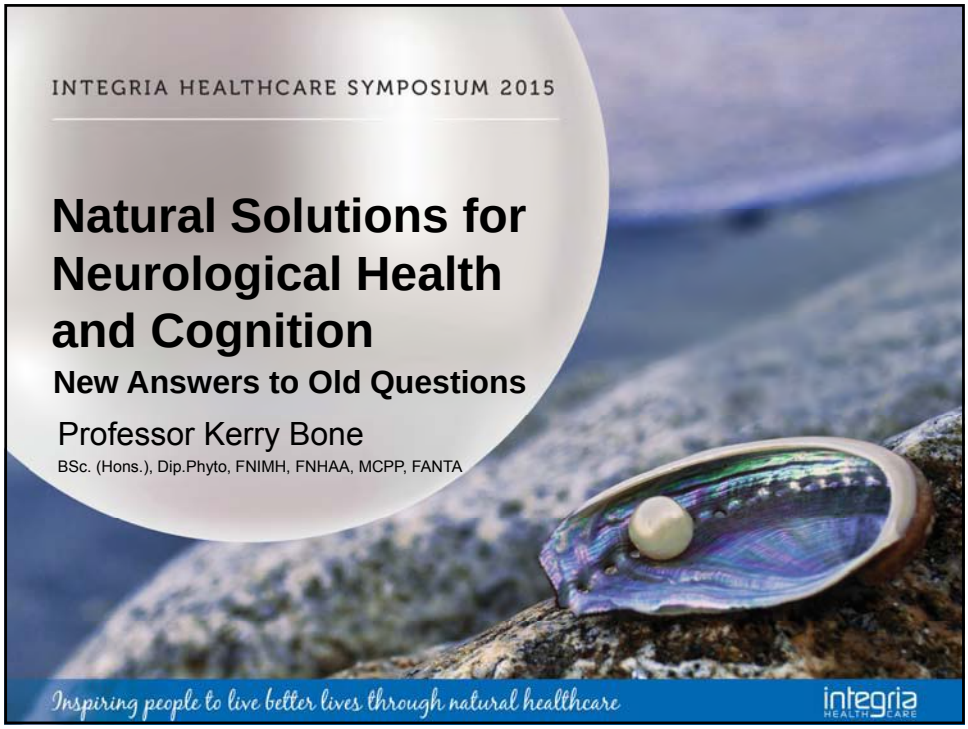
DAY 2



Professor Kerry Bone
BSc. (Hons.), Dip.Phyto, FNIMH, FNHAA, MCPP, FANTA

Inspiring people to live better lives through natural healthcare

integria HEALTH CARE



Methodology

- More than 2500 titles reviewed
- Around 700 abstracts selected and read
- Around 200 papers read, mostly reviews

Optimising Brain Health

- Behind most dysfunctional brain disorders are:
  - Neuroinflammation (NI)
  - Oxidative and nitrosative stress ⇒
  - Mitochondrial dysfunction
  - AND Neurotransmitter dysregulation (where relevant)

 3

Optimising Brain Health

- This includes key disorders such as:
  - Alzheimer's disease (AD) and age-related cognitive decline (CD)
  - Depression/bipolar disorder
  - Schizophrenia
  - Attention deficit hyperactivity disorder (ADHD) and autism
  - Central sensitivity syndrome (CSS)

 4

Immuno-exitotoxicity

- First coined by Blaylock in 2008
- Cytokines and glutamate receptors interact to ↑↑↑ sensitivity of glutamate receptor system
- Central is chronic activation of brain's innate immune system (mainly microglial cells)

Blaylock RL, Maroon J. *Surg Neurol Int* 2012; **3**: 19

 5

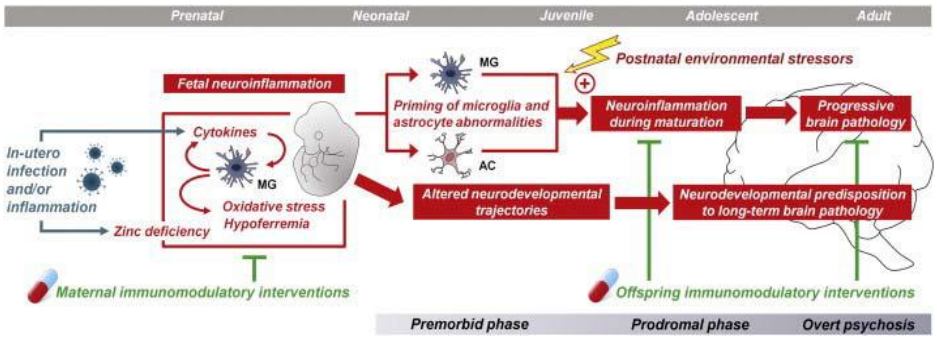
Immuno-exitotoxicity

- Hence excitotoxicity can occur even at low levels of extracellular glutamate
- As pathology develops CNS ⇒ even more vulnerable ⇒ loss of antioxidant systems (including SOD and glutathione)
- ⇒ Mitochondrial energy loss

Blaylock RL, Maroon J. *Surg Neurol Int* 2012; **3**: 19

 6

Schizophrenia and NI

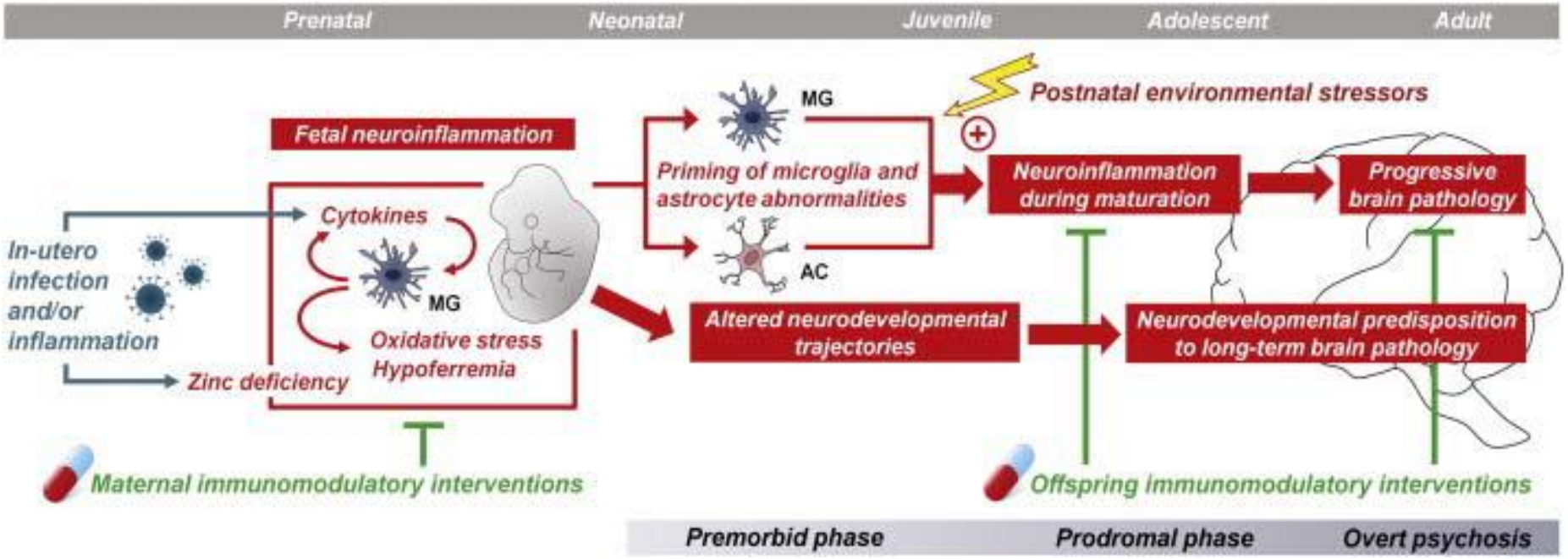


Meyer U. *Neuropsychopharmacol Biol Psychiatry* 2013; **42**: 20-34

Core Strategy

- Downregulate neuroinflammation
- Support brain mitochondrial function
- Upregulate antioxidant protection
- Enhance neuroprotection/cytoprotection

Schizophrenia and NI



Meyer U. *Neuropsychopharmacol Biol Psychiatry* 2013; **42**: 20-34

Brain and Immunity

- Brain has unique immunophysiology: 'immune-privileged'
- Innate immune system ⇒ fights infections, even in late stages
- ↓ Acquired immune response ⇒ long-term persistence of inflammatory responses in brain

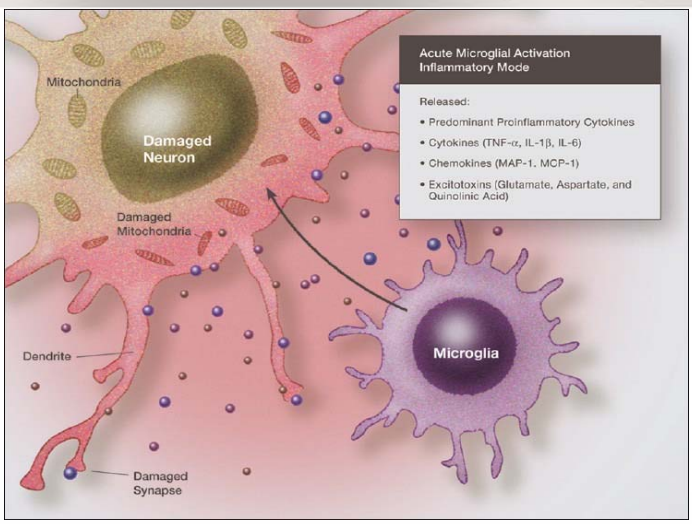
Arnett SV, Alleva LM, Korossy-Horwood R et al. *Med Hypotheses* 2011; **77**(1): 77-83

Brain and Immunity

- Transport proteins in blood-brain barrier (BBB) ⇒ facilitate systemic cytokines into brain
- Net result: systemic inflammation ⇒ persistent inflammatory response within brain

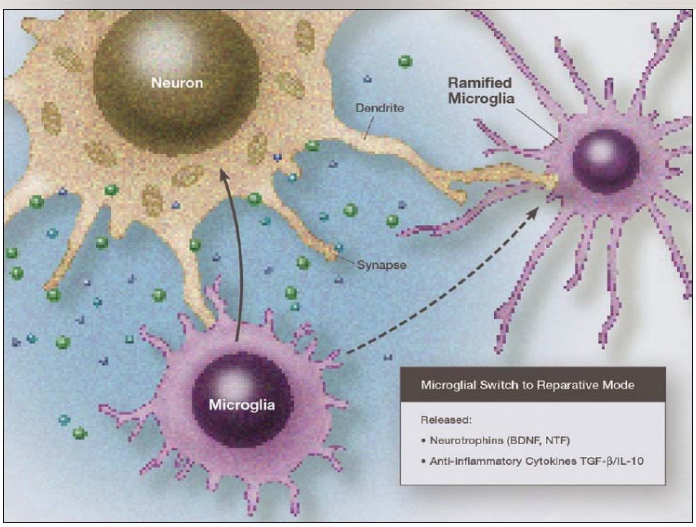
Arnett SV, Alleva LM, Korossy-Horwood R et al. *Med Hypotheses* 2011; **77**(1): 77-83

Brain and Immunity



Blaylock RL. *Surg Neurol Int* 2013 Sep; **4**: 118

Brain and Immunity



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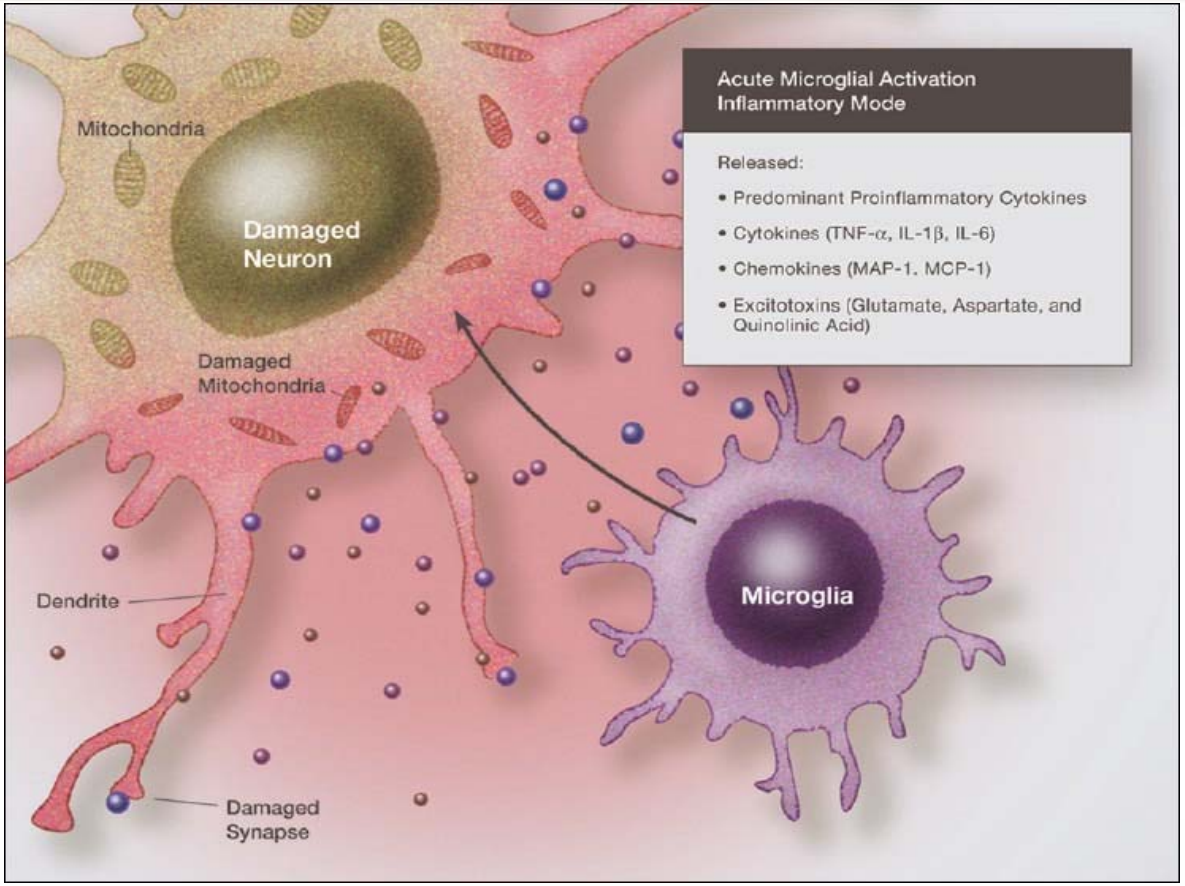
Brain and Immunity



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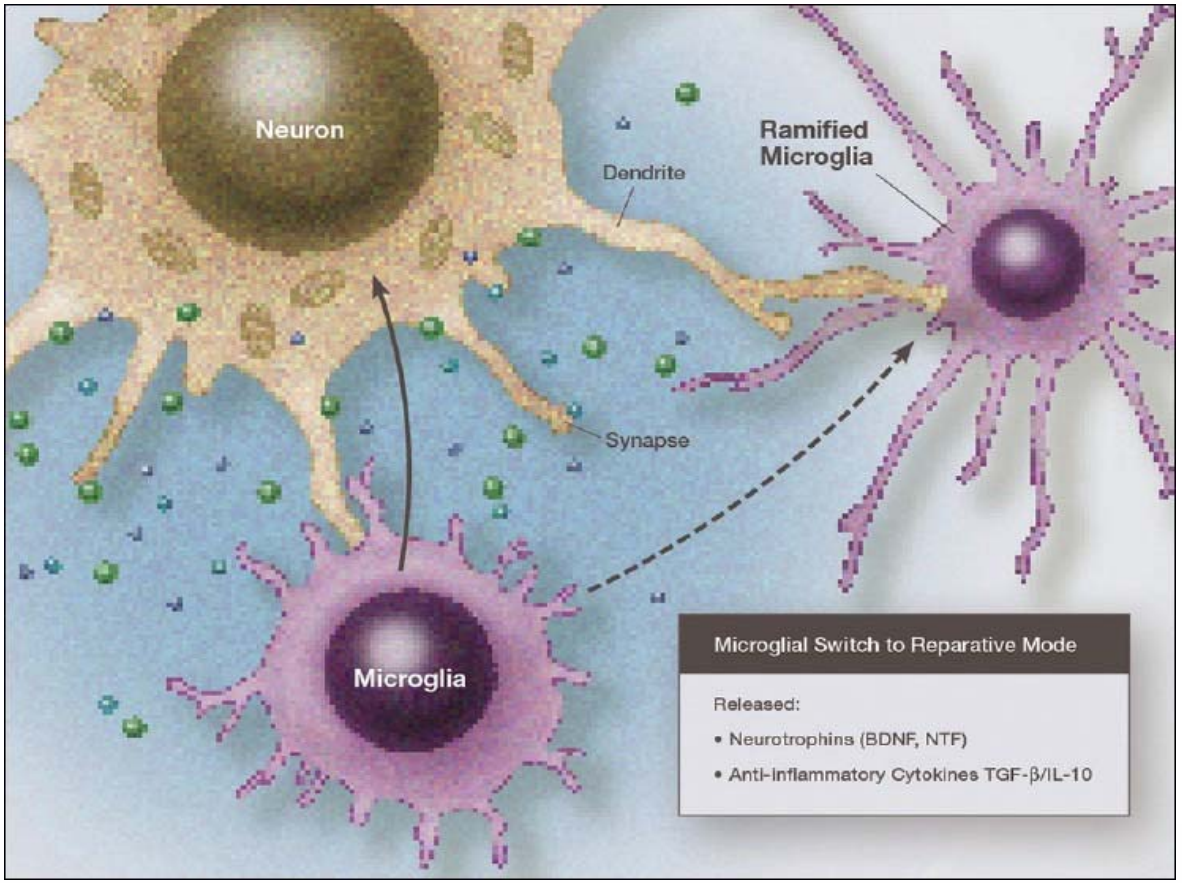

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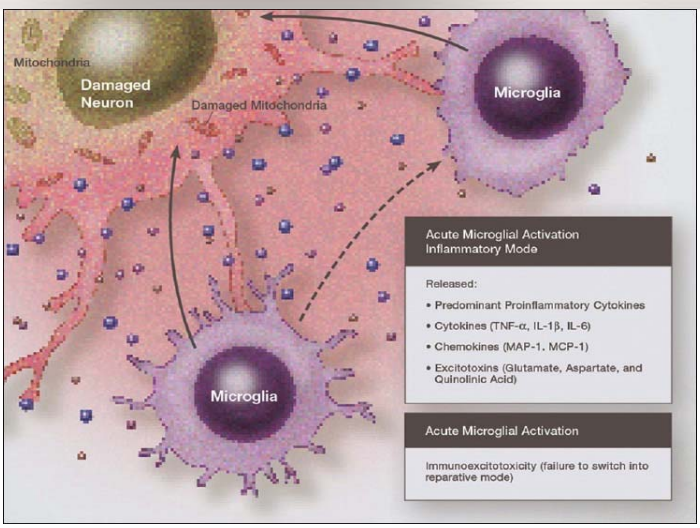
Brain and Immunity



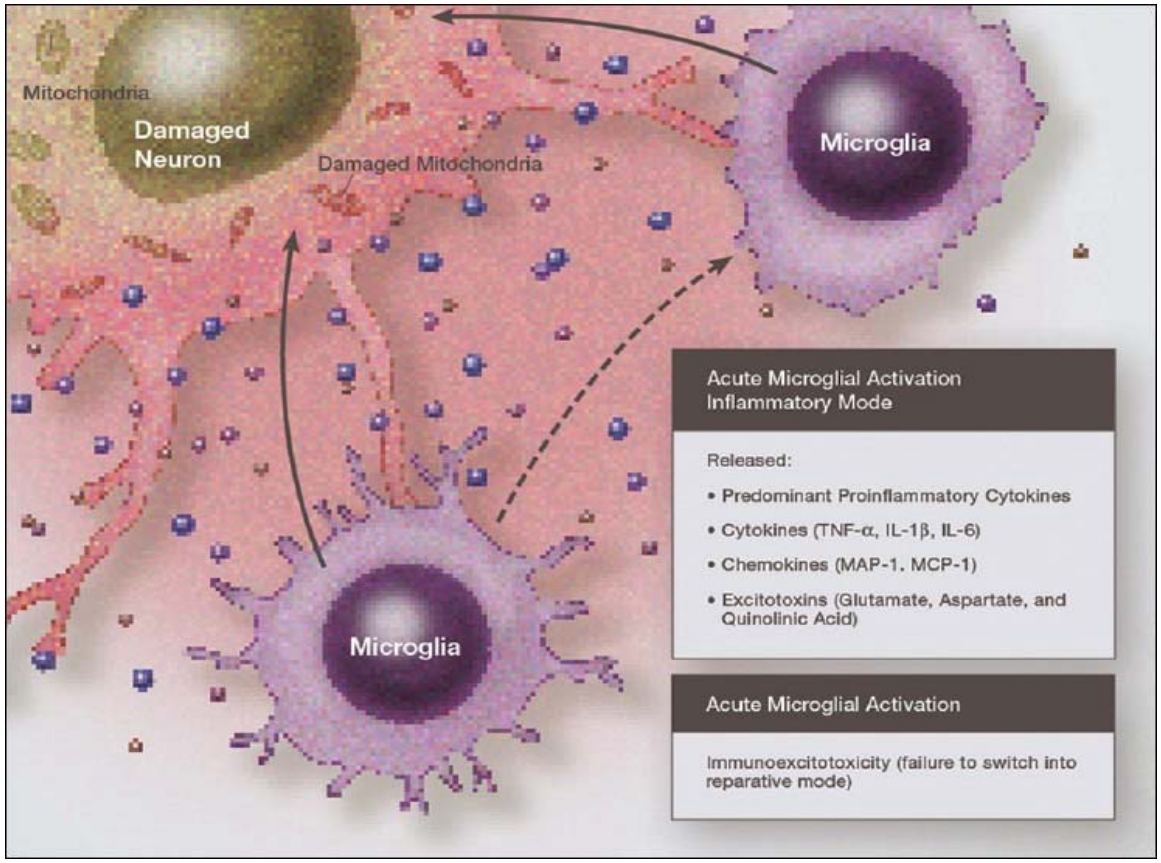
Blaylock RL. *Surg Neurol Int* 2013 Sep; **4**: 118

How Microglia Are Activated

- Receptors, eg pattern recognition receptors (PPRs) such as Toll-like receptors (TLRs)
- Elevated levels of oxidative and nitrosative stress (O & NS)
- Chemokines and cytokines

Blaylock RL. *Surg Neurol Int* 2013 Sep; **4**: 118

Brain and Immunity



Blaylock RL. *Surg Neurol Int* 2013 Sep; **4**: 118

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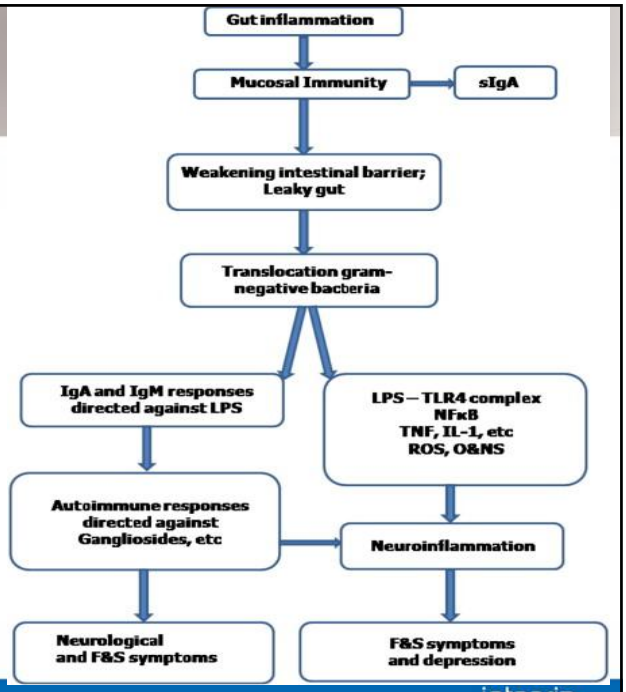
Switching Off NI

- ↓ Systemic inflammation and its drivers
- ↓ Brain inflammation, eg Boswellia, curcumin and omega-3 fatty acids

Drivers of Systemic Inflammation

- Stealth pathogens and occult viruses
- Chronic infections
- Autoimmunity and other chronic inflammatory conditions
- Dysbiosis and bacterial translocation
- Diet

NI Drivers

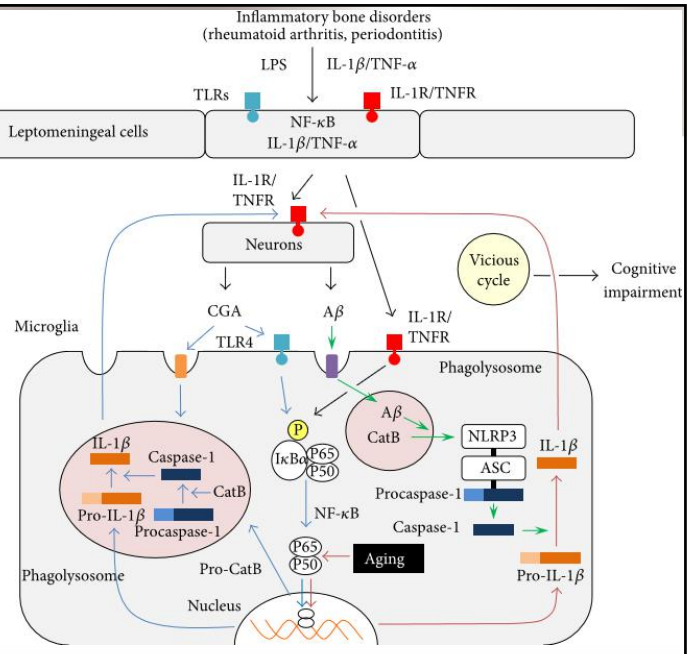


Maes M, Twisk FN, Kubera M et al. *J Affect Disord* 2012; **136**(3) :909-917

Drivers of Systemic Inflammation

- Stress and a dysregulated HPA axis function
- O and NS
- Fatty liver and insulin resistance
- Trauma
- Drugs

Chronic Infection and AD



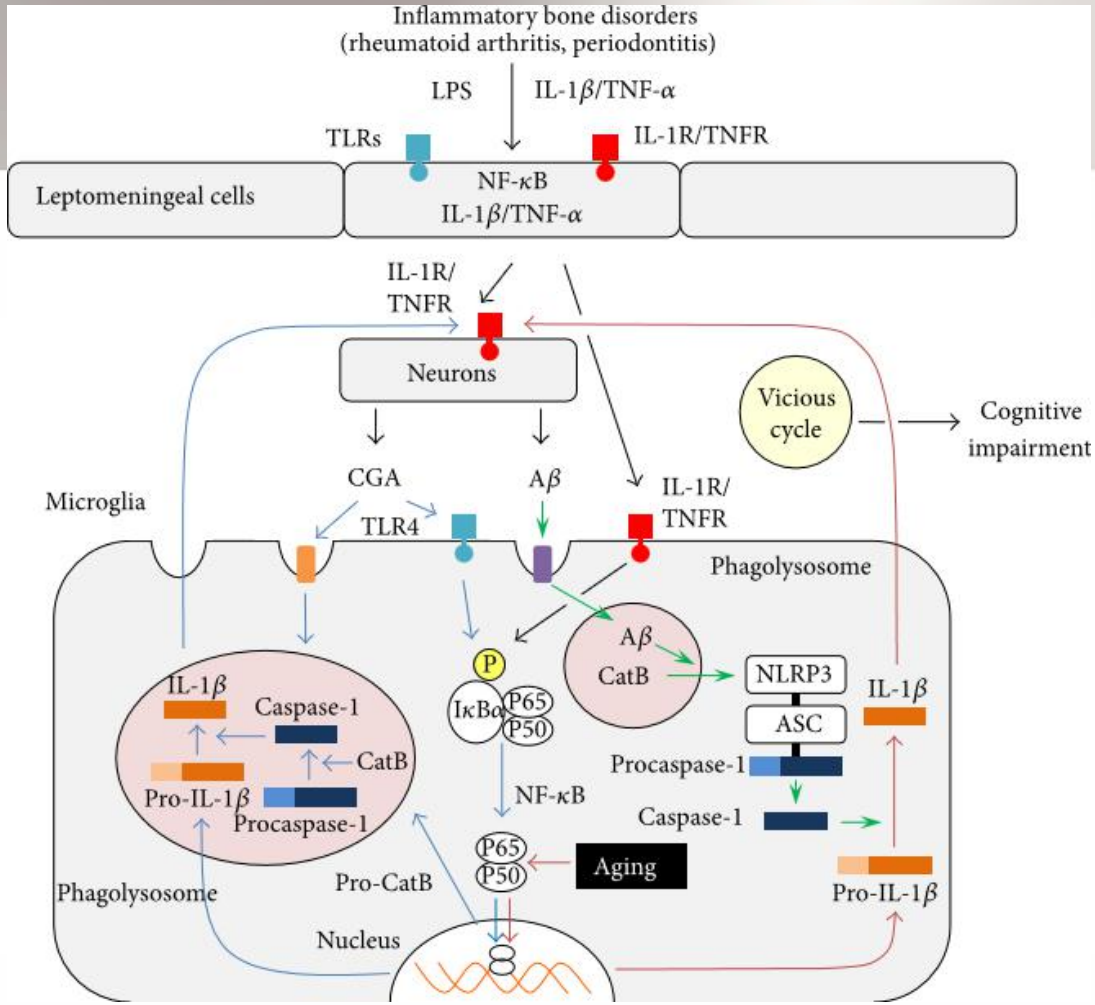
Wu Z, Nakanishi H. *J Pharmacol Sci* 2014; **126**(1): 8-13

Stealth Pathogens and AD

- After 20 years of follow up ⇒
  - 28.9% of *Helicobacter pylori* positive subjects developed dementia
  - versus 21.1% of negative subjects
  - p = 0.04

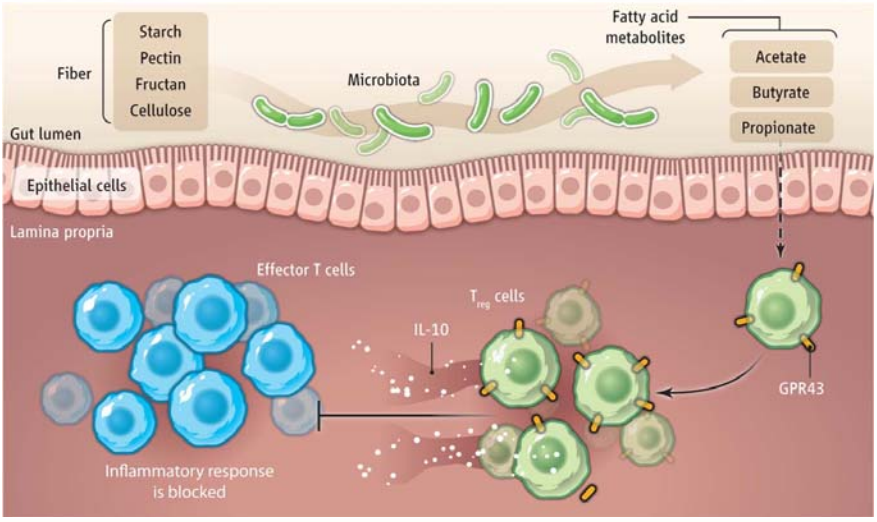
Adriani A, Fagoonee S, De Angelis C et al. *Panminerva Med* 2014 Sep; **56**(3): 195-199

Chronic Infection and AD



Wu Z, Nakanishi H. *J Pharmacol Sci* 2014; **126**(1): 8-13

Gut Health and Inflammation



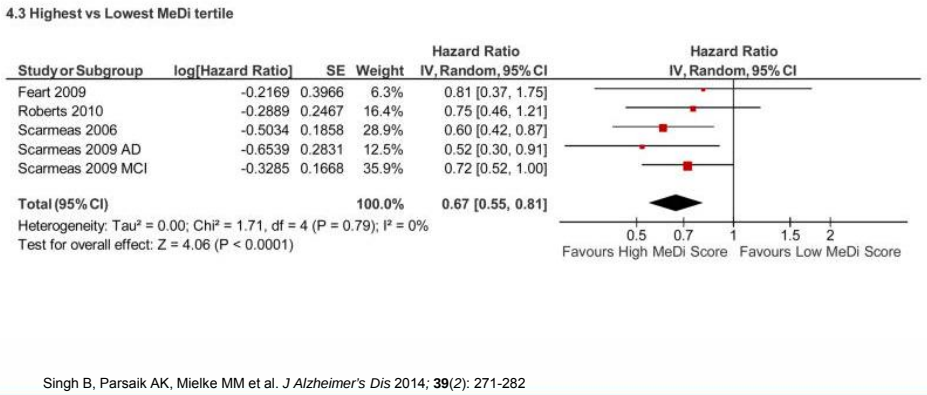
Anti-inflammatory Diet

- ↓ Fried, baked etc foods (↓ dietary AGEs)
- ↑ Fibre – grains, seeds, fruit, vegetables, pulses etc
- ↑ Fats (omega-3); ↓ fats (omega-6), ↑ neutral fats (monounsaturates)

Anti-inflammatory Diet

- ↑ Flavonoids and other key phytochemicals (fruit, vegetables, spices, herbs, cocoa etc)
- ↓ Fructose
- Fluid pH balance in body tissues : ↑ alkalinity by limiting animal protein
- Free from provocative antigens and potential toxins

Mediterranean Diet and CD



Singh B, Parsaik AK, Mielke MM et al. *J Alzheimer's Dis* 2014; **39**(2): 271-282

MIND Diet and CD

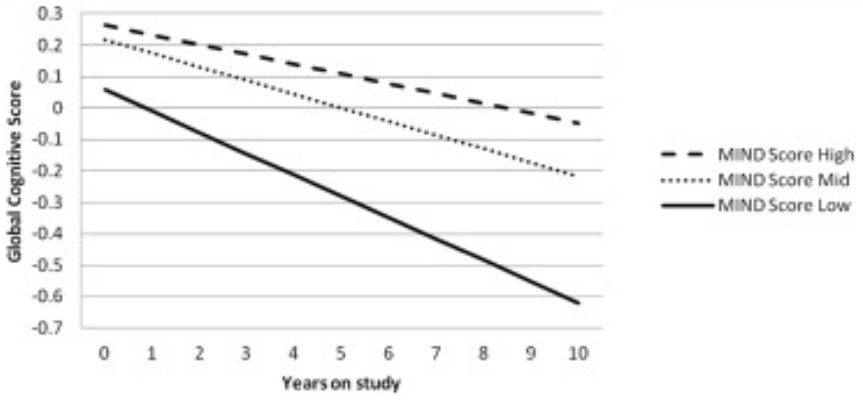
MIND Diet Component Servings and Scoring			
Diet Component	**0**	**0.5**	**1**
Green Leafy Vegetables	≤ 2 serving/wk	> 2 to < 6/wk	≥ 6 servings/wk
Other Vegetables	< 5 serving/wk	5 to < 7 wk	≥ 1 servings/d
Berries	< 1 serving/wk	1/wk	≥ 2 serving/wk
Nuts	< 1/mo	< 1/mo to < 5/wk	≥ 5 servings/wk
Olive oil	Not primary oil		Primary oil used
Butter	> 2 T/d	1-2/d	< 1 T/d
Cheese	7 + servings/wk	1-6/wk	< 1 serving/wk
Whole Grains	< 1 serving/d	1-2/d	≥ 3 servings/d

MIND Diet and CD

MIND Diet Component Servings and Scoring			
Diet Component	**0**	**0.5**	**1**
Fish (not fried)	Rarely	1-3/mo	≥ 1 meals/wk
Beans	<1 meal/wk	1-3/wk	> 3 meals/wk
Poultry (not fried)	< 1 meal/wk	1/wk	≥ 2 meals/wk
Red Meat and products	7 + meals/wk	4-6/wk	< 4 meals/wk
Fast fried foods	4 + times/wk	1-3/wk	< 1 time/wk
Pastries and sweets	7 + servings/wk	5-6/wk	< 5 servings/wk
Wine	>1 glass/d	1/mo-6/wk	1 glass/d
Total Score			15

Morris MC, Tangney CC, Wang Y et al. *Alzheimers Dement* 2015 Jun 15; [Epub ahead of print]

MIND Diet and CD



Morris MC, Tangney CC, Wang Y et al. *Alzheimers Dement* 2015; [Epub ahead of print]

Dietary AGEs and CD

- Observational study in healthy adults (n=93) ≥ 60 years of age[1]
- Over 9 months
- High baseline methyl-glyoxal derivatives (MGs) in blood predicted cognitive decline (p = 0.041)

1. Cai W, Uribarri J, Zhu L et al. *Proc Natl Acad Sci U S A* 2014; **111**(13): 4940-4945

Dietary AGEs and CD

- Deterioration in insulin sensitivity also correlated with MGs[1]
- Reinterpretation of existing epidemiological studies ⇒ dietary AGEs appear to be important risk factors for AD[2]

1. Cai W, Uribarri J, Zhu L et al. *Proc Natl Acad Sci U S A* 2014; **111**(13): 4940-4945
2. Perrone L, Grant WB. *J Alzheimers Dis* 2015; **45**(3): 965-979

 29

Core Strategy

- Downregulate neuroinflammation
- Support brain mitochondrial function
- Upregulate antioxidant protection
- Enhance neuroprotection/cytoprotection

 30

Support Mitochondrial Function

- Magnesium
- Acetyl-L-carnitine (ALC)
- N acetylcysteine (NAC) and Nrf2 herbs
- Ubiquinol
- Alpha lipoic acid (ALA)

Support Mitochondrial Function

- Omega-3 fatty acids
- Ribose
- Vitamin B co-factors (including B6, folate, B12)
- Medium chain fatty acids (MCFA)
- Key herbs: Hawthorn, resveratrol (Polygonum) and Ginkgo

Coconut Oil

Table 1. Fatty acid composition of coconut oil, showing percentage of total fat

Name	% Total fat	Saturated/ unsaturated	MCFA/ LCFA
Caproic acid (6:0)	0·6	Saturated	MCFA
Caprylic (8:0)	0·8	Saturated	MCFA
Capric (10:0)	6·4	Saturated	MCFA
Lauric (12:0)	48·5	Saturated	MCFA
Myristic (14:0)	17·6	Saturated	MCFA
Palmitic acid (16:0)	8·4	Saturated	MCFA
Stearic acid (18:0)	2·5	Saturated	LCFA
Linoleic acid (18:1)	6·5	Unsaturated	LCFA
Linolenic (18:2)	1·5	Unsaturated	LCFA

MCFA, medium-chain fatty acids; LCFA, long-chain fatty acids.

Fernando WM, Martins IJ, Goozee KG et al. *Br J Nutr* 2015; **114**(1): 1-14

MCFA and Mitochondria

- MCFA readily enter mitochondria independently of carnitine transport system
- Readily converted to ketone bodies and further metabolised
- Ketogenic diet enhances brain mitochondrial ATP production

Fernando WM, Martins IJ, Goozee KG et al. *Br J Nutr* 2015; **114**(1): 1-14

Neuroprotection/Cytoprotection

- Key neuroprotective herbs: Ginkgo and Saffron; and nutrients: NAC, B12 and ALA
- ↓ O and NS via Nrf2 herbs: Ginkgo, Broccoli sprouts, Turmeric, Rosemary, Green Tea etc
- ↑ HSPs: adaptogens?, Echinacea root?
- ↑ SIRT1: Polygonum (resveratrol)
- ↑ Microcirculation protection: Gotu Kola, Grape Seed, Ginkgo and 5-Point Plan

Reiterating……

- AD and other neurodegenerative disorders
- Parkinson's disease (PD)
- Schizophrenia
- Autism
- Depression (including bipolar disorder)
- ADHD
- Central sensitivity syndrome

AD Risk Factors

Adverse Risk Factors

	AD	CVD
Midlife hypertension	✓	✓
Diabetes	✓	✓
Hyperlipidaemia	✓(midlife)	✓
Smoking, current	✓	✓
Head injury	✓	×
Obesity	✓(midlife)	✓
Age	✓	✓
Homocysteine	✓	✓

CVD = cardiovascular disease

Hughes TM, Craft S, Lopez OL. *Neurodegener Dis Manag* 2015; **5**(2): 121-135
Imtiaz B, Tolppanen AM, Kivipelto M et al. *Biochem Pharmacol* 2014; **88**(4): 661-670

AD Risk Factors

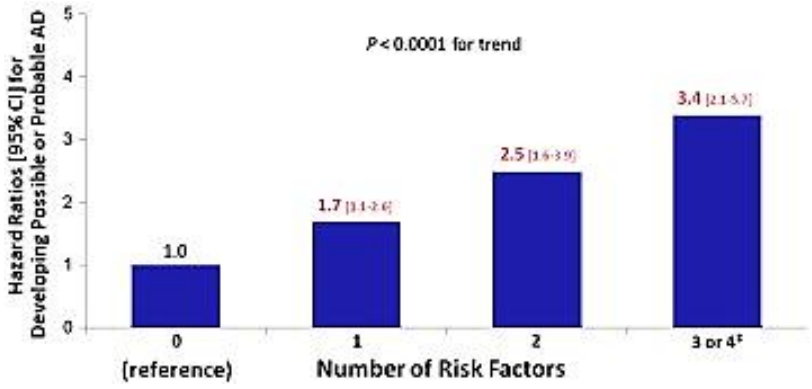
Protective Risk Factors

	AD	CVD
Exercise	✓	✓
Education > 15 years	✓	×
NSAID use	✓	× (adverse)

NSAID = non-steroidal anti-inflammatory drugs

Hughes TM, Craft S, Lopez OL. *Neurodegener Dis Manag* 2015; **5**(2): 121-135
Imtiaz B, Tolppanen AM, Kivipelto M et al. *Biochem Pharmacol* 2014; **88**(4): 661-670

AD Risk Factors

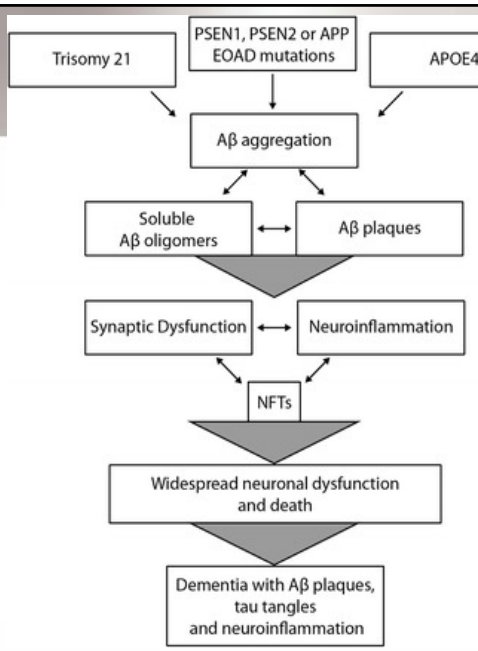


Kalaria RN, Akinyemi R, Ihara M. *J Neurol Sci* 2012; **322**(1-2): 141-147

AD Causal Theories

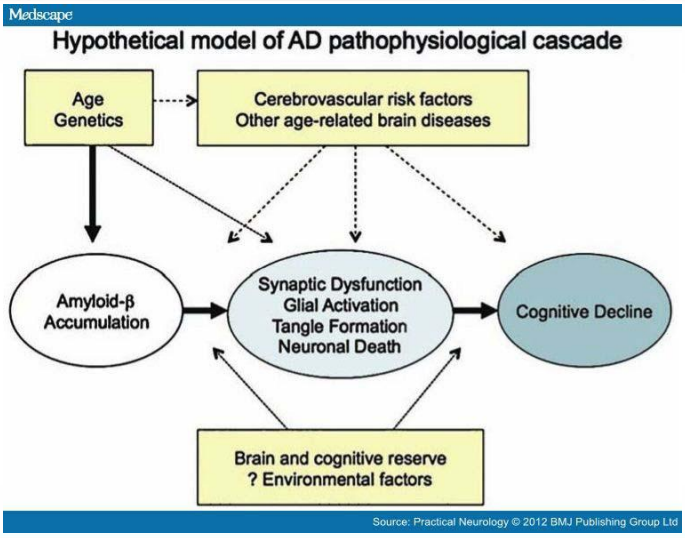
- Neuritic plaques (amyloid beta, A_β) and neurofibrillary tangles (tau)
- Neuroinflammatory
- Vascular
- Weird and wonderful

The Amyloid Hypothesis

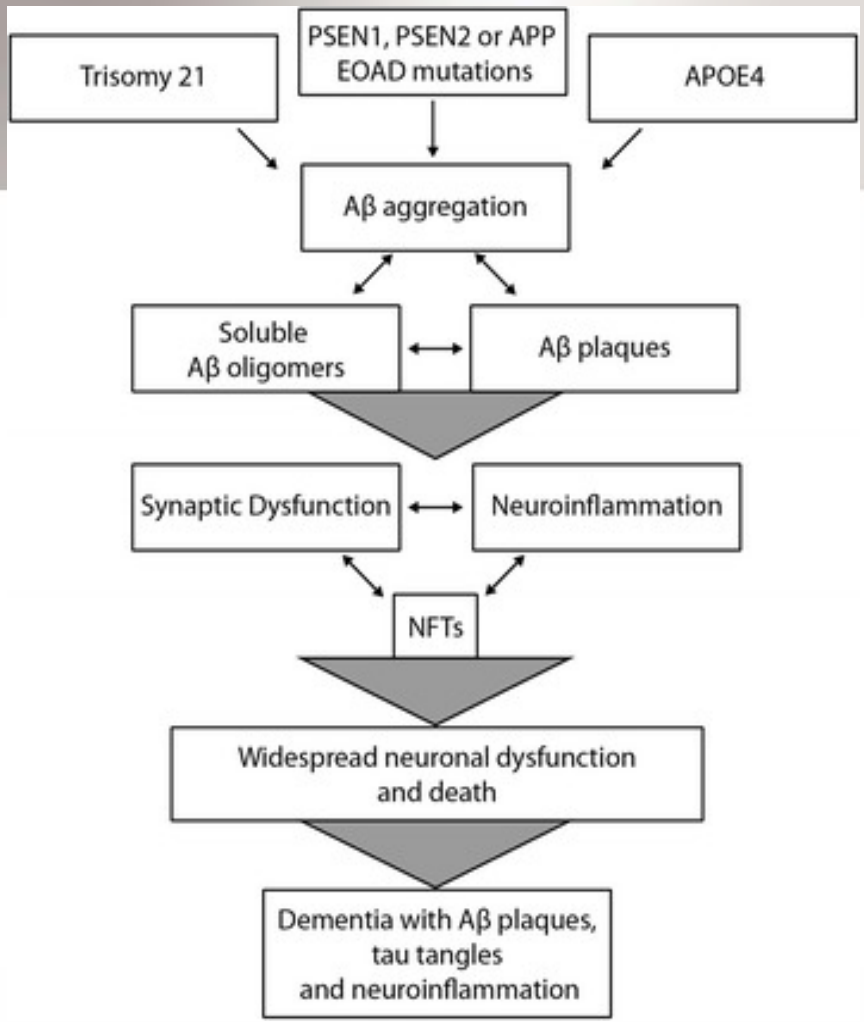


Morris GP, Clark IA, Vissel B. *Acta Neuropathol Commun* 2014; **2**: 135

The Amyloid Hypothesis

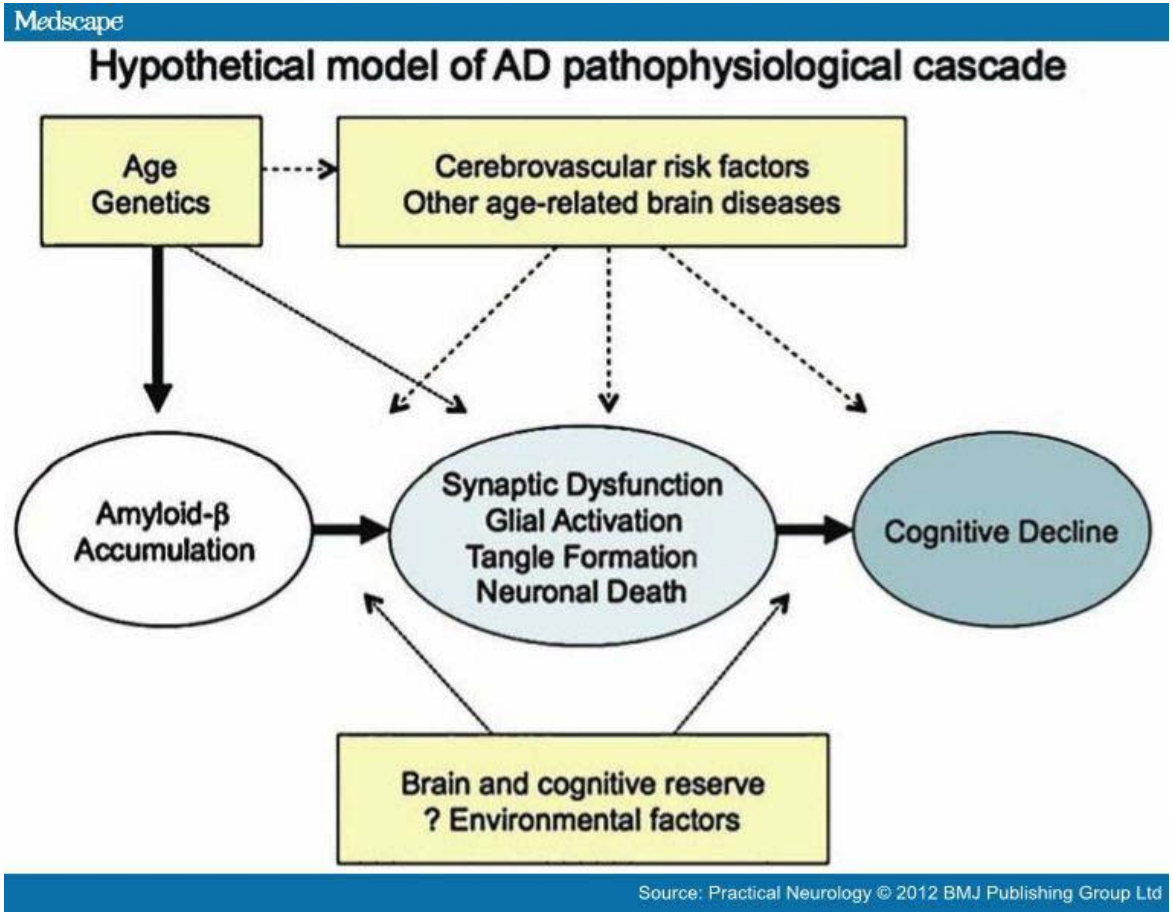


The Amyloid Hypothesis

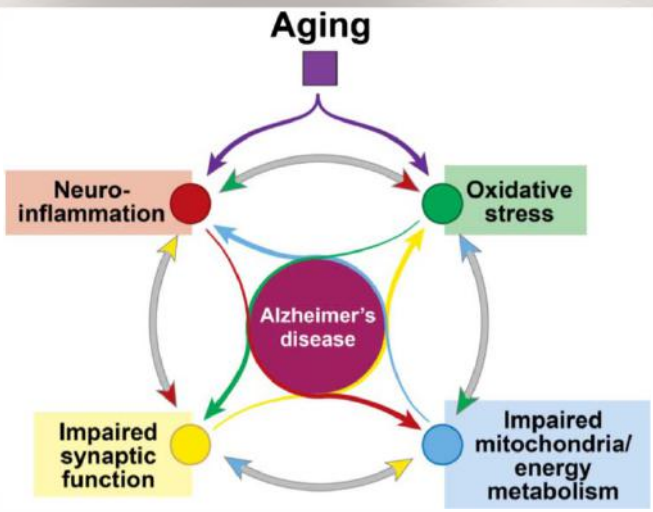


Morris GP, Clark IA, Vissel B. *Acta Neuropathol Commun* 2014; **2**: 135

The Amyloid Hypothesis

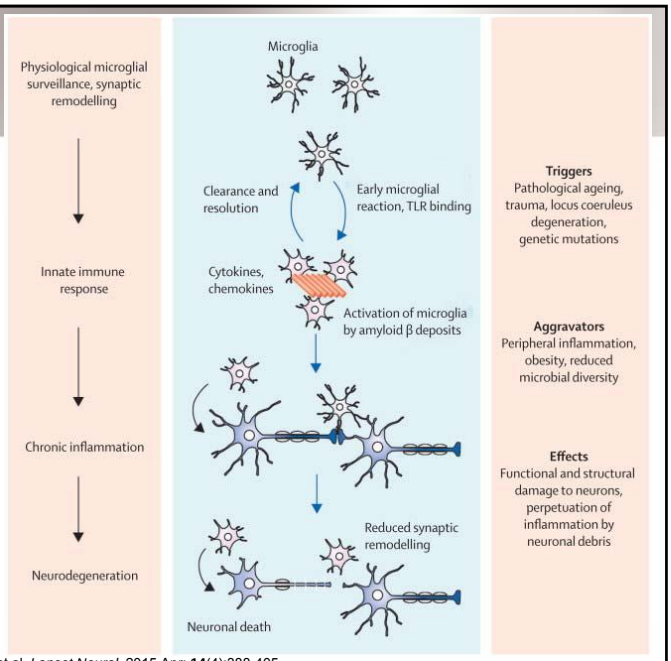


Neuroinflammation and AD



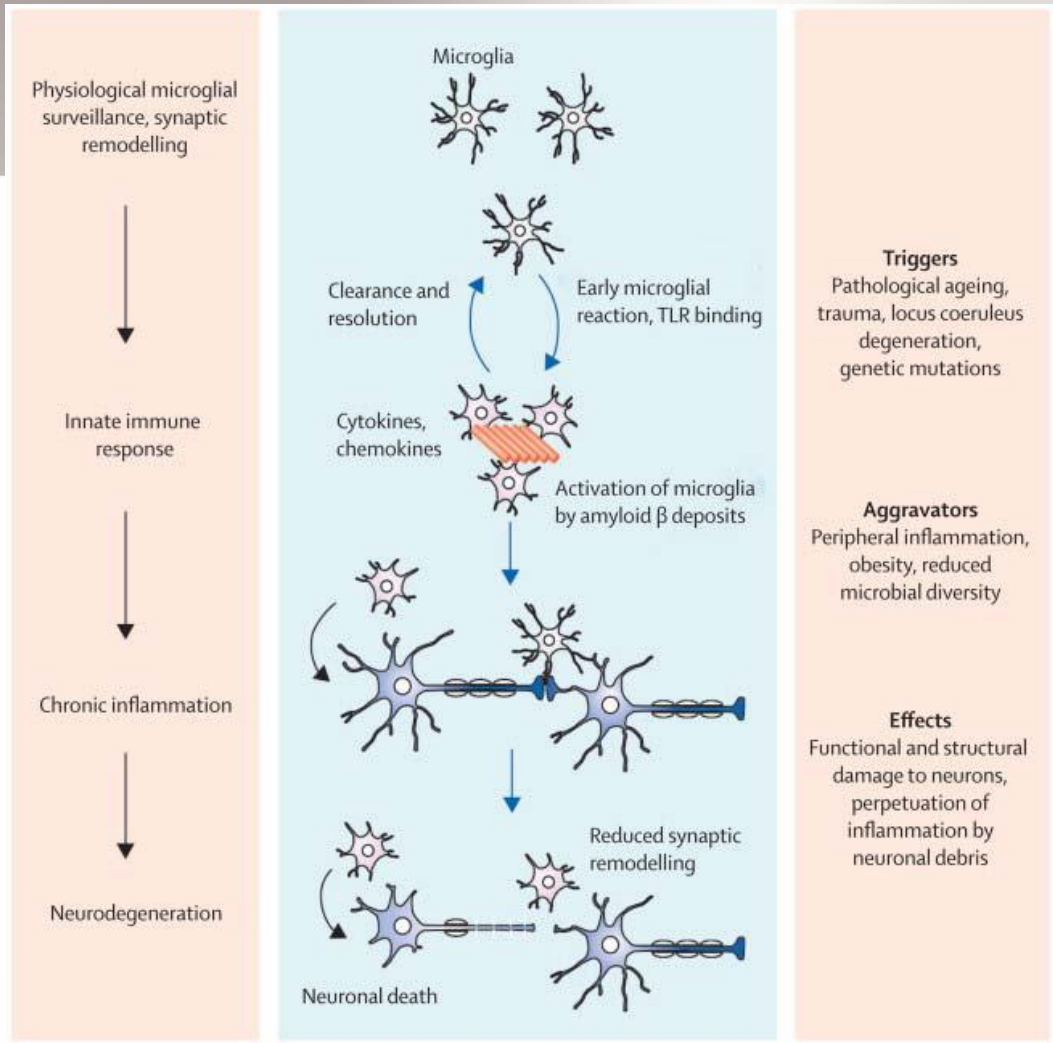
Pimplikar SW. *J Clin Immunol* 2014; **34** (Suppl 1): S64-9

NI and AD

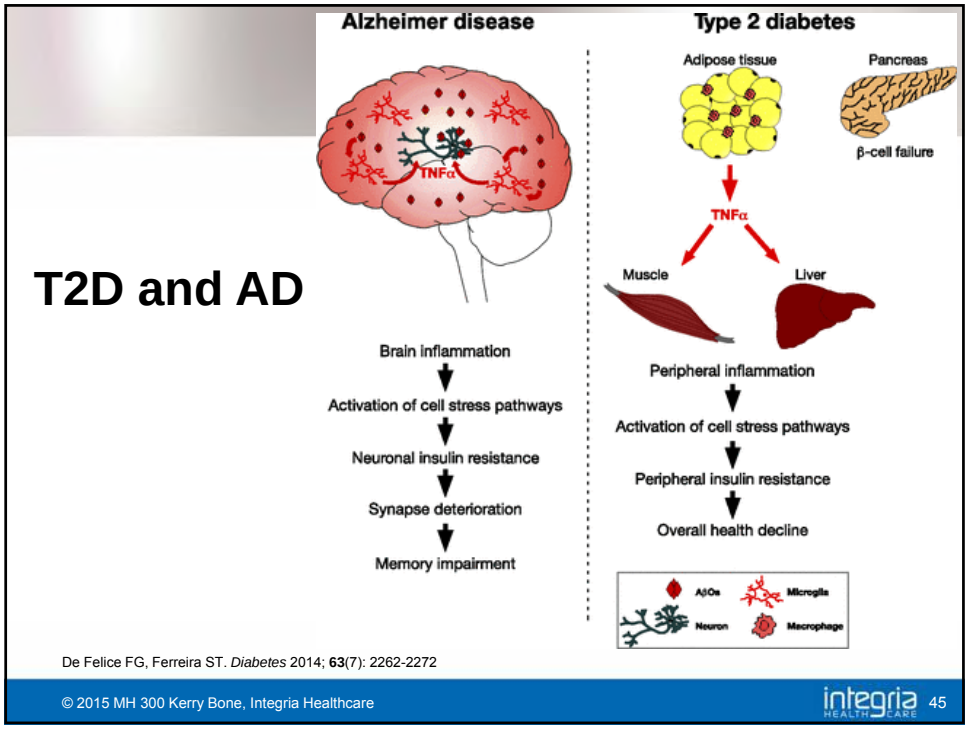


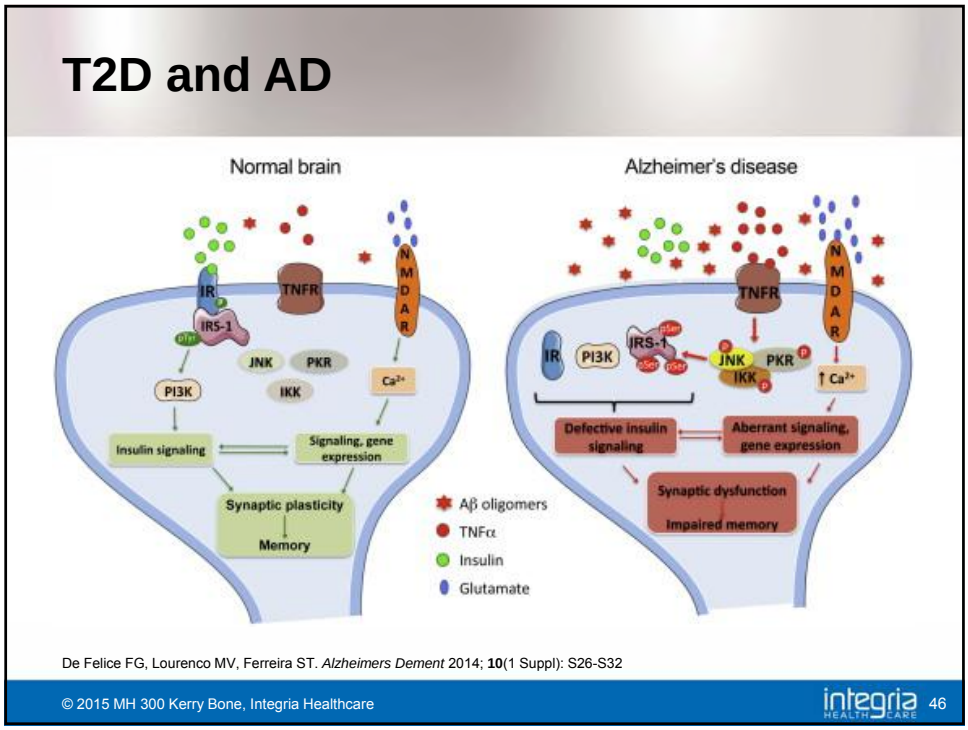
Heneka MT, Carson MJ, El Khoury J et al. *Lancet Neurol.* 2015 Apr; **14**(4):388-405

NI and AD

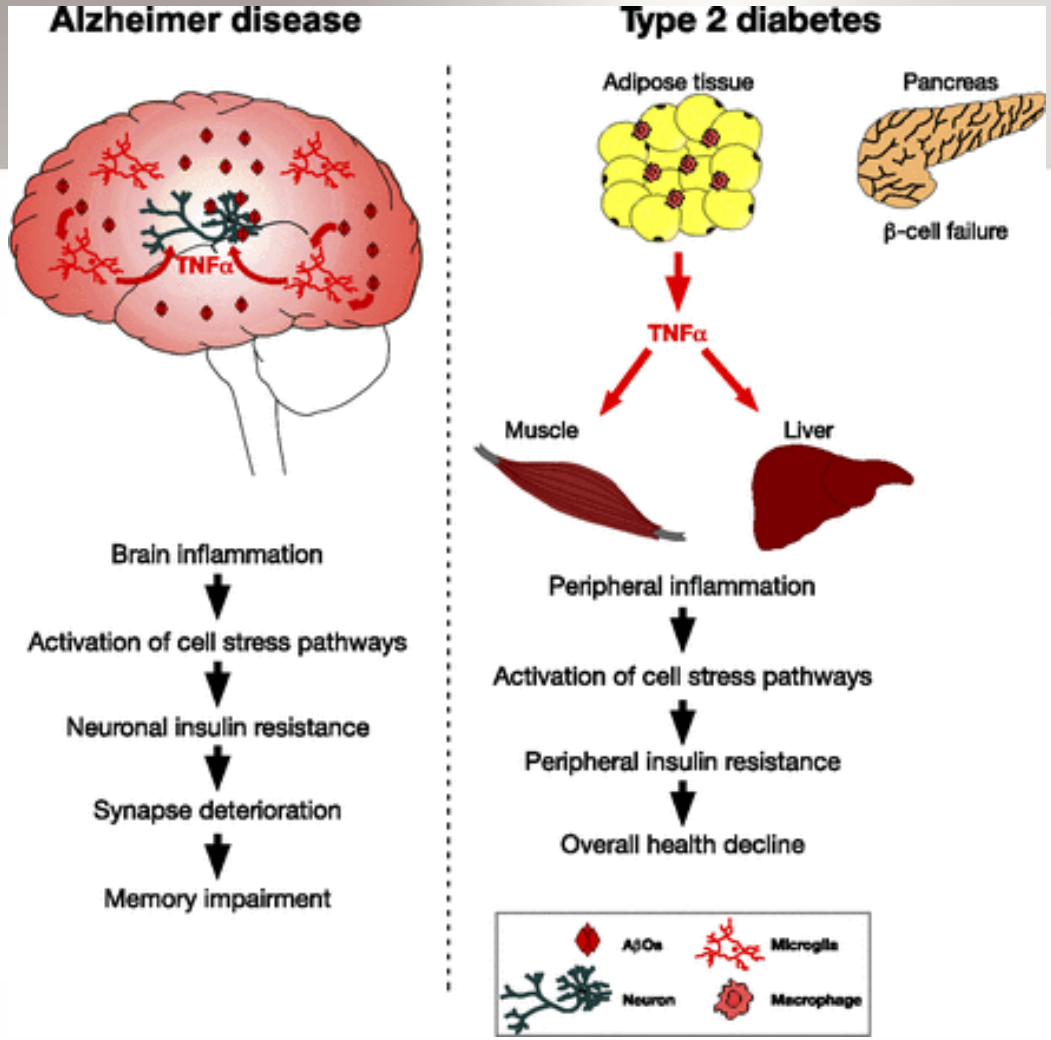


Heneka MT, Carson MJ, El Khoury J et al. *Lancet Neurol.* 2015 Apr; **14**(4):388-405



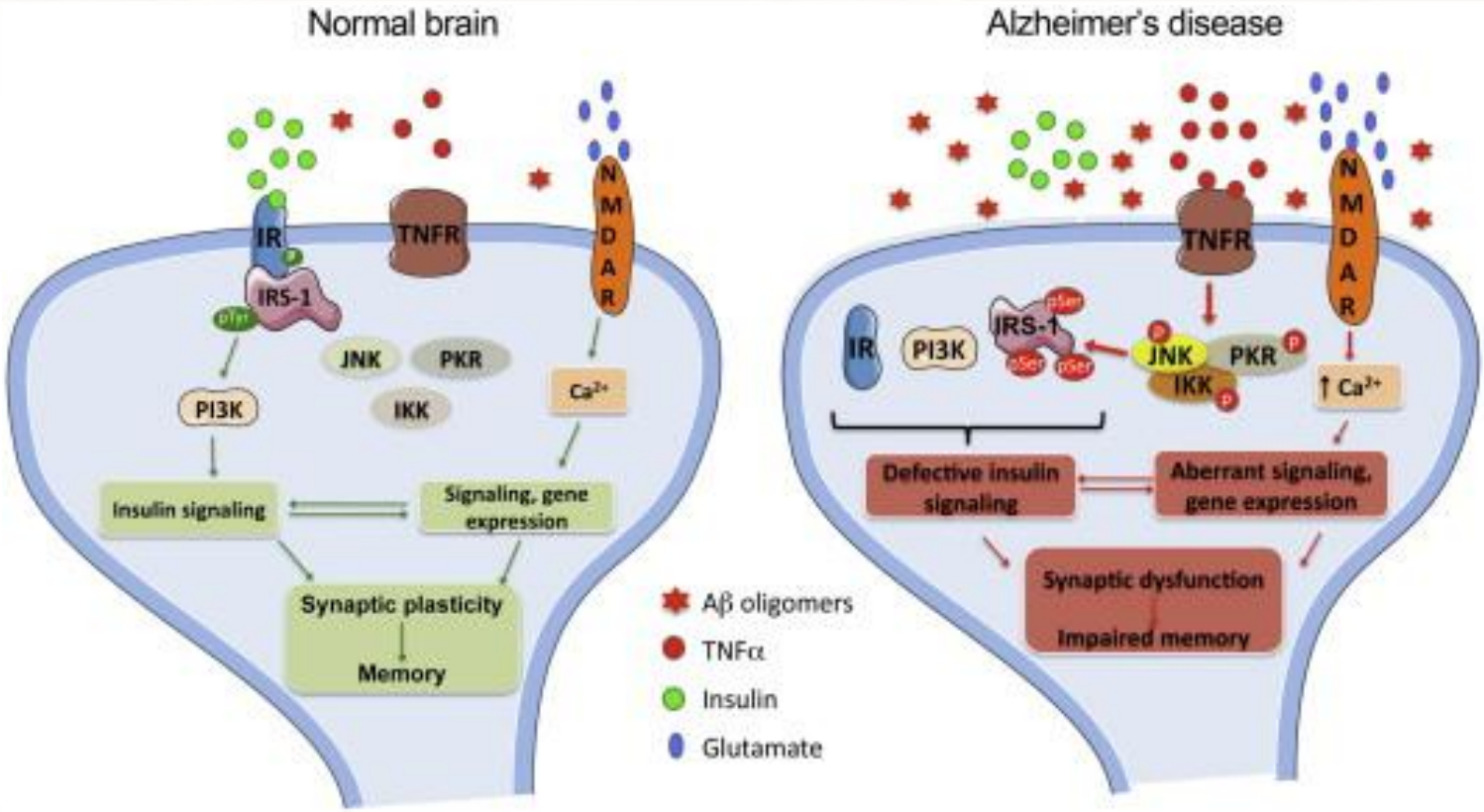


T2D and AD



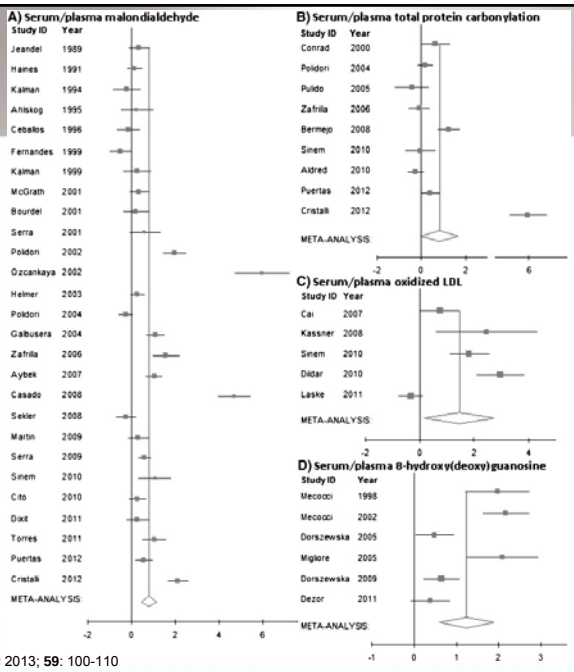
De Felice FG, Ferreira ST. *Diabetes* 2014; **63**(7): 2262-2272

T2D and AD



De Felice FG, Lourenco MV, Ferreira ST. *Alzheimers Dement* 2014; **10**(1 Suppl): S26-S32

O & NS in AD



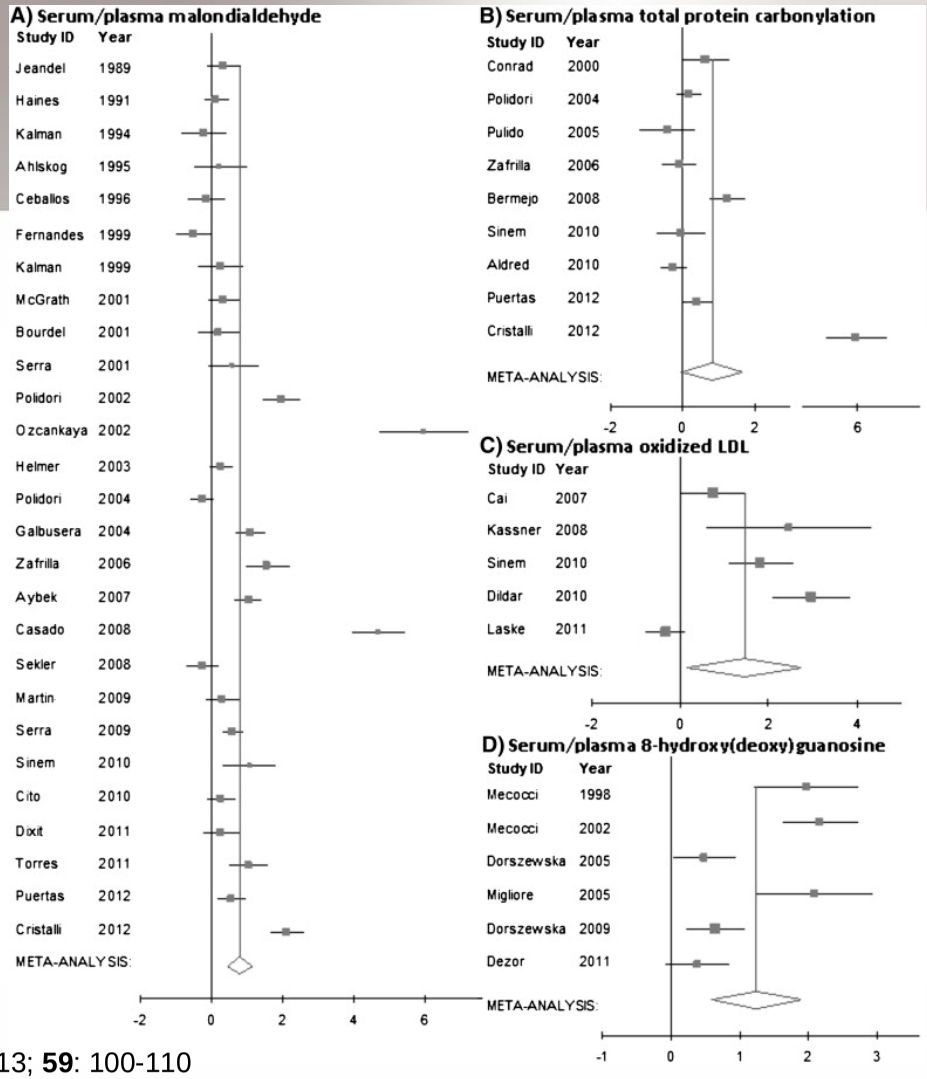
Schrag M, Mueller C, Zabel M et al. *Neurobiol Dis* 2013; **59**: 100-110

Mitochondria and AD

- Large body of evidence ⇒ mitochondrial dysfunction plays a central role in AD
- Not just isolated to neurons
- Mitochondrial dysfunction ↑ with ageing
- Transmission of AD risk preferentially found in maternal inheritance

Morris JK, Honea RA, Vidoni ED et al. *Biochim Biophys Acta* 2014; **1842**(9): 1340-1349

O & NS in AD



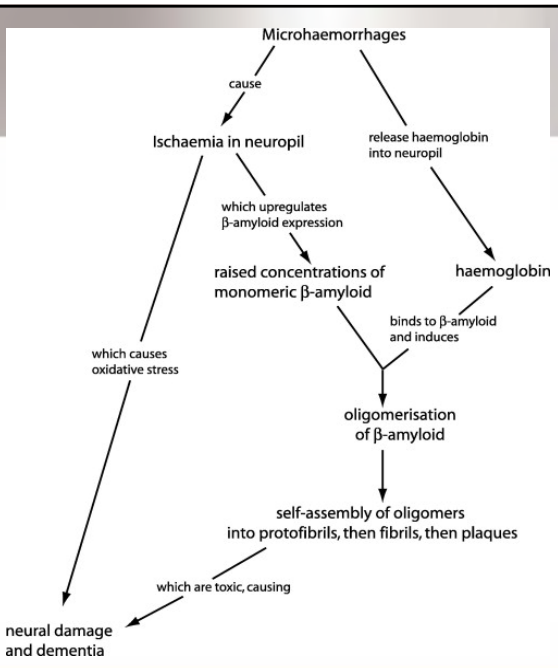
Schrag M, Mueller C, Zabel M et al. *Neurobiol Dis* 2013; **59**: 100-110

Vascular Theory of AD

"The beat of our heart is symbolic of life, of energy, of courage and determination. Yet… if we live to old age, the heart destroys us. And it does so in a terrible way, pummelling the brain beat after beat until its small blood vessels burst, and lesions, tens of thousands of them, erode its circuitry, until the brain shrinks around the debris, its function failing. Slowly, relentlessly – this evidence goes - the beat of the heart destroys the memory, the intellect and the personality of the person it had so long served to keep alive." Jonathan Stone

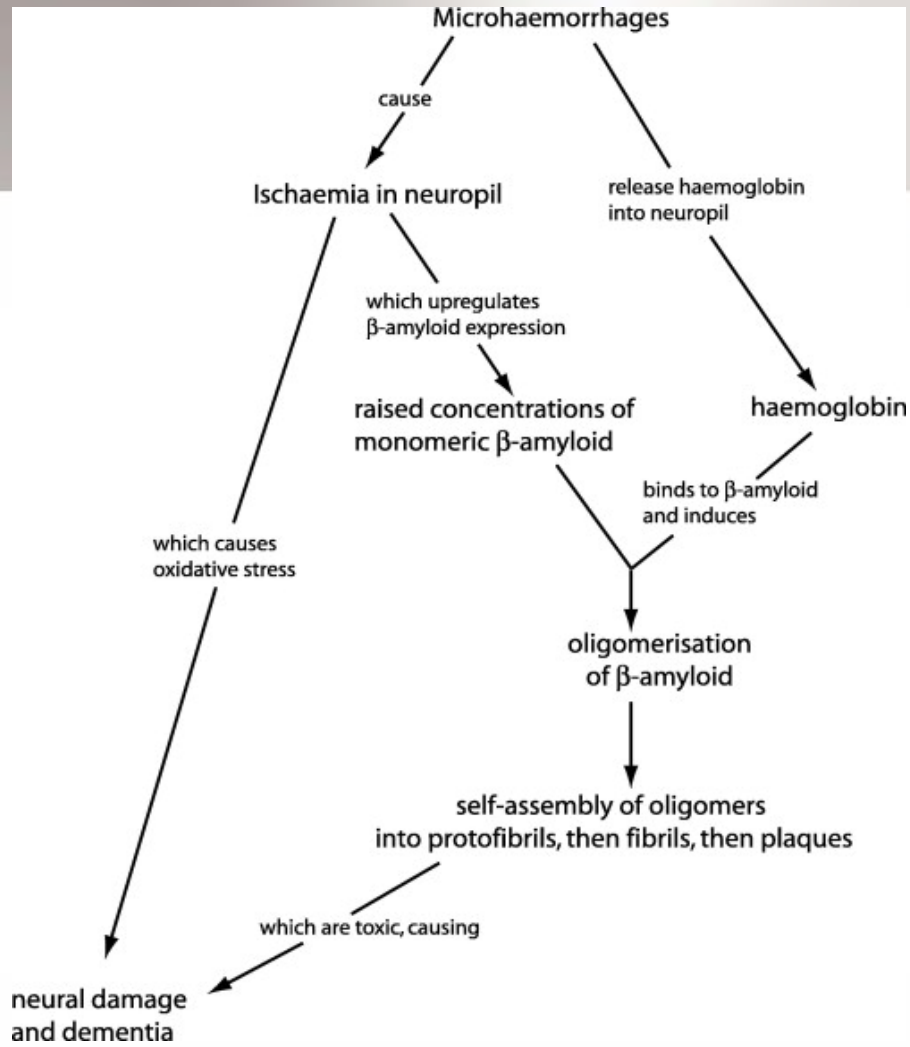
http://www.abc.net.au/radionational/programs/ockhamsrazor/dementia3a-a-tale-of-two-organs/6051492#transcript

Vascular Theory of AD



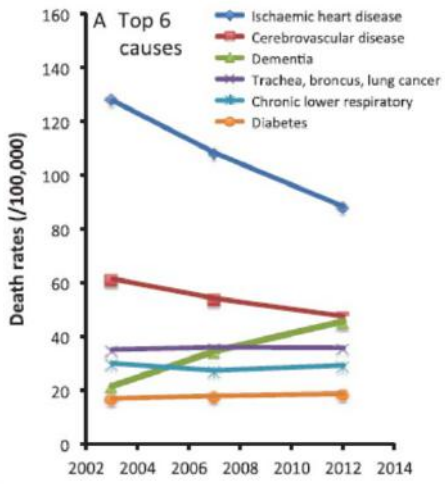
Stone J. *Med Hypotheses* 2008; **71**(3): 347-359

Vascular Theory of AD



Stone J. *Med Hypotheses* 2008; **71**(3): 347-359

Causes of Death



Stone J, Johnstone DM, Mitrofanis J et al. *J Alzheimers Dis* 2015; **44**(2): 355-373

Arterial Stiffness

- Markedly higher in AD
- Ten of twelve studies ⇒ ↑ vascular stiffness ⇒ impaired cognition
- Caused by:
  - loss of elastic components (eg aortic elastin) with ageing
  - atherosclerosis and resultant calcification

Çalık AN, Özcan KS, Yüksel G et al. *Clin Interv Aging* 2014; **9**: 1115-1121
Hughes TM, Craft S, Lopez OL. *Neurodegener Dis Manag* 2015; **5**(2): 121-135
Rabkin SW. *J Alzheimers Dis* 2012; **32**(3): 541-549

Does Paracetamol Cause AD?

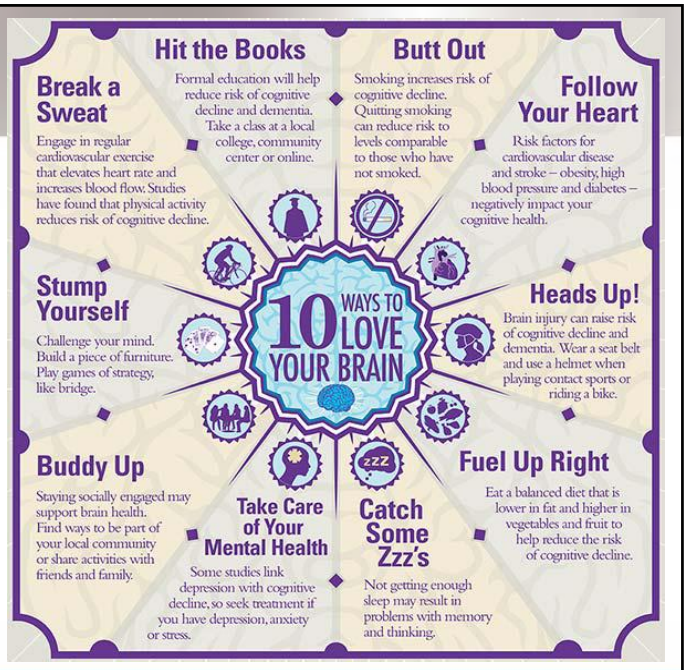
- Introduction of phenacetin (1887) loosely correlates to abrupt emergence of AD
- Nephrotoxicity ⇒ phenacetin metabolite paracetamol (PA)
- Not well studied, but PA use linked to AD risk
- PA could exacerbate/cause neuroinflammation
- Antidotes would be NAC and tyrosine

Jones GR. *Inflamm Allergy Drug Targets* 2014 Feb;**13**(1): 2-14

Preventing AD

- Develops slowly ⇒ when symptoms appear is difficult to reverse
- Is a progressive disease ⇒ so what might ↓ risk will also ↓ progression

Preventing AD



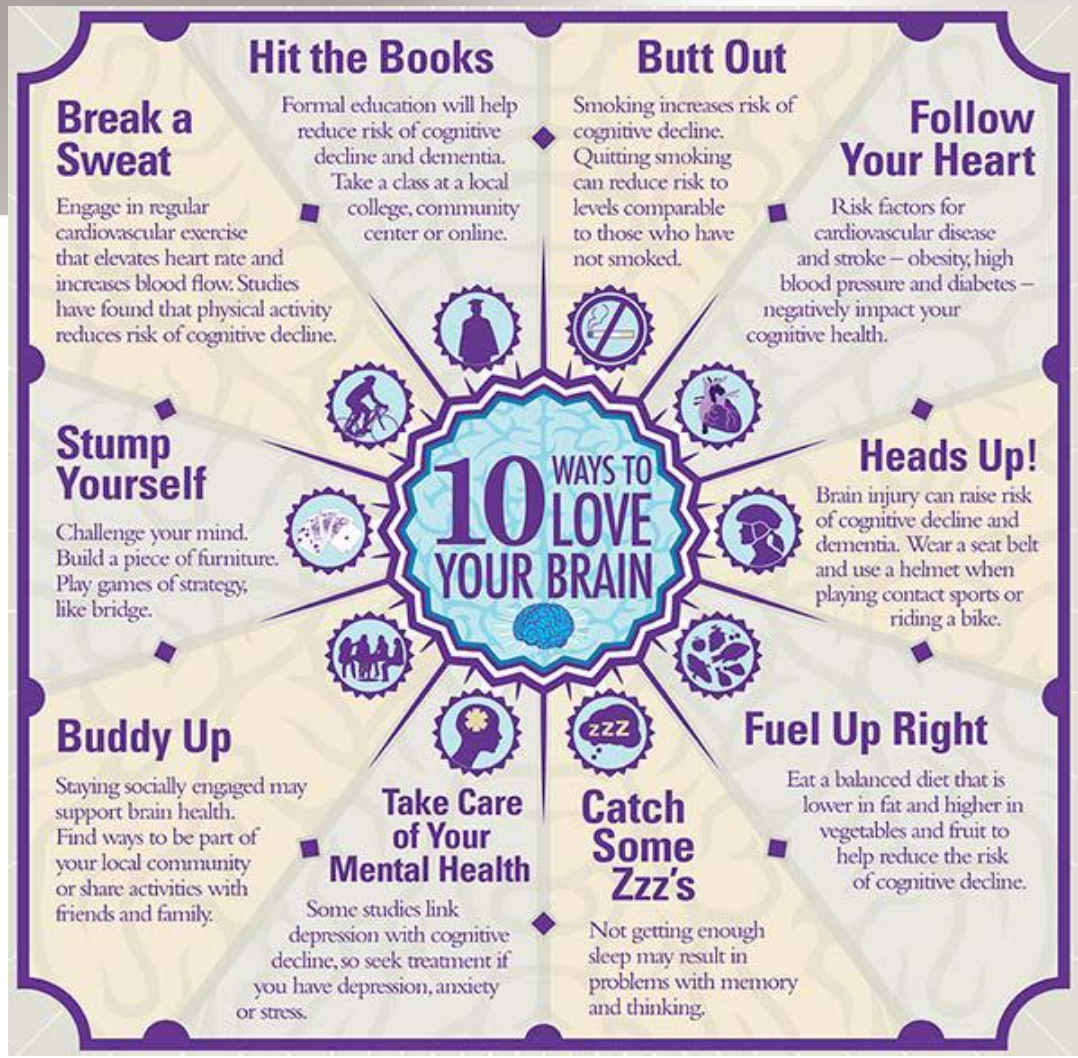
 55

Key Strategies in Prevention

- ↓ Neuroinflammation
- ↓ O and NS
- ↑ Mitochondrial support
- ↑ Neuroprotection/cytoprotection
- ↑ Microcirculatory health and arterial flexibility

 56

Preventing AD



Key Herbs and Supplements

- Ginkgo
- Saffron, Turmeric (curcumin), adaptogens, resveratrol
- ALC and NAC
- Omega-3 fatty acids
- Vitamin B12 and adequate folate

Dietary and Lifestyle Aspects

- As per slide 55
- A diet that scores at least 10 on the MIND scale
- Medium chain fatty acids (coconut)
- The 5-Point Microcirculation Dietary Plan

The 5-Point Dietary Plan

1. Boost dietary nitrate
2. Increase cocoa intake
3. Increase berry anthocyanin intake
4. Raw crushed garlic: ½ to 1 clove/day
5. Increase herbs and spices: green tea, turmeric and ginger

Treating AD

- Treatment goals same as for prevention (slide 56)
- In addition: be guided by the case to address specific drivers of systemic inflammation
- Dietary and lifestyle goals as per prevention (slides 55 and 58)

Key Herbs and Supplements

- Ginkgo, Saffron, Bacopa and Sage
- Bioavailable curcumin, Boswellia, resveratrol
- NAC and ALC

Key Herbs and Supplements

- ALA, Mg, Zn, vitamin B12 and ubiquinol
- Omega-3 fatty acids
- Treatments to address neuroinflammation drivers (slides 15 to 20)

Ginkgo and AD

- Canadian 2014 meta-analysis: Ginkgo extract (120 to 240 mg/day) effective and safe in dementia[1]
- Chinese 2015 meta-analysis: Ginkgo extract (240 mg/day) effective in cognitive impairment and dementia (including AD sub-group)[2]

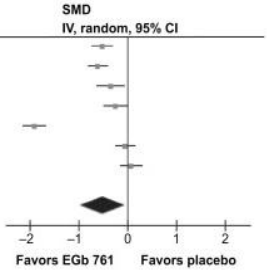
1. Gauthier S, Schlaefke S. *Clin Interv Aging* 2014; **9**: 2065-2077
2. Tan MS, Yu JT, Tan CC et al. *J Alzheimers Dis* 2015; **43**(2): 589-603

Ginkgo and AD

A

Study or subgroup	EGb 761 Mean	SD	Total	Placebo Mean	SD	Total	Weight	SMD IV, random, 95% CI
Herrschaft et al[13]	−2.2	3.5	200	−0.3	3.7	202	14.4%	−0.53 (−0.73, −0.33)
Ihl et al[36]	−1.4	2.8	202	0.3	2.7	202	14.4%	−0.62 (−0.82, −0.42)
Kanowski et al[33,34]	−2.1	3.1	105	−1	3.1	98	14.0%	−0.35 (−0.63, −0.08)
Le Bars et al[30,31]	−0.3	5.35	136	1	5.32	134	14.2%	−0.24 (−0.48, −0.00)
Napryeyenko et al[35]	−3.2	2.3	198	1.3	2.4	197	14.2%	−1.91 (−2.15, −1.67)
Nikolova et al[14]	−2.2	3.6	196	−2	4	201	14.4%	−0.05 (−0.25, 0.14)
Schneider et al[32]	1.3	5.5	170	0.9	5.6	174	14.3%	0.07 (−0.14, 0.28)
Total (95% CI)			**1,207**			**1,208**	**100.0%**	**−0.52 (−0.98, −0.05)**

Heterogeneity: Tau²=0.38; Chi²=189.27, *df*=6 (*P*<0.00001); *F*=97%
Test for overall effect: *Z*=2.18 (*P*=0.03)



Gauthier S, Schlaefke S. *Clin Interv Aging* 2014; **9**: 2065-2077

Saffron and AD

- Saffron (30 mg/day stigma extract) versus donepezil (10 mg/day) in 54 patients with moderate AD ⇒ equal efficacy after 22 weeks[1]
- More effective than placebo (same dose) in 46 AD patients over 16 weeks[2]
- Recently, saffron (same dose) versus memantine (20 mg/day) in 68 patients with moderate to severe AD ⇒ equal efficacy after 12 months[3]

1. Akhondzadeh S, Shafiee Sabet M, Harirchian MH et al. *Psychopharmacology (Berl)* 2010; **207**(4): 637-643
2. Akhondzadeh S, Sabet MS, Harirchian MH et al. *J Clin Pharm Ther* 2010; **35**(5): 581-588
3. Farokhnia M, Shafiee Sabet M, Iranpour N et al. *Hum Psychopharmacol* 2014; **29**(4): 351-359

Saffron and Retinal Flicker

- Retinal flicker sensitivity (RFS) ⇒ dynamic retinal vascular diameter change in response to flickering light ⇒ endothelial/microvascular function[1]
- RFS ↓ in AD patients[2]
- Saffron (20 mg/day) improved RFS in an RCT of 25 patients with early macular degeneration[3]

1. Chang LY, Lowe J, Ardiles A et al. *Alzheimers Dement* 2014; **10**(2): 251-261
2. http://www.abc.net.au/worldtoday/content/2015/s4217520.htm
3. Falsini B, Piccardi M, Minnella A et al. *Invest Ophthalmol Vis Sci* 2010; **51**(12): 6118-6124

NAC and AD

- Recent systematic review found two uncontrolled reports and one 24-week randomised, controlled trial (RCT)
- RCT showed improvement in some, but not all cognitive testing
- Rated evidence as C

Deepmala, Slattery J, Kumar N et al. *Neurosci Biobehav Rev* 2015; **55**: 294-321

 67

ALC and AD

- ALC has been described as a mitochondrial rejuvenator[1]
- Meta-analysis of 21 RCTs of ALC in mild cognitive impairment or mild AD ⇒ significant effect over placebo (most trials 1.5 to 2.0 g ALC/day)[2]

1. Rosca MG, Lemieux H, Hoppel CL. *Adv Drug Deliv Rev* 2009; **61**(14): 1332-1342
2. Montgomery SA, Thal LJ, Amrein R. *Int Clin Psychopharmacol* 2003; **18**(2): 61-71

 68

VITACOG Study

- 266 participants with mild cognitive impairment (>70 years) treated for 2 years
- Homocysteine ↓ 30%
- B vitamins ↓ cognitive decline, especially if homocysteine is elevated
- Brain atrophy↓

de Jager CA, Oulhaj A, Jacoby R et al. *Int J Geriatr Psychiatry* 2012; **27**(6): 592-600
Smith AD, Smith SM, de Jager CA et al. *PLoS One* 2010; **5**(9): e12244

Chicago Health and Ageing Project

- Higher serum methylmalonic acid (MMA) ⇒ faster rate of cognitive decline
- Higher serum B12 ⇒ slower rate
- Homocysteine ⇒ no relationship

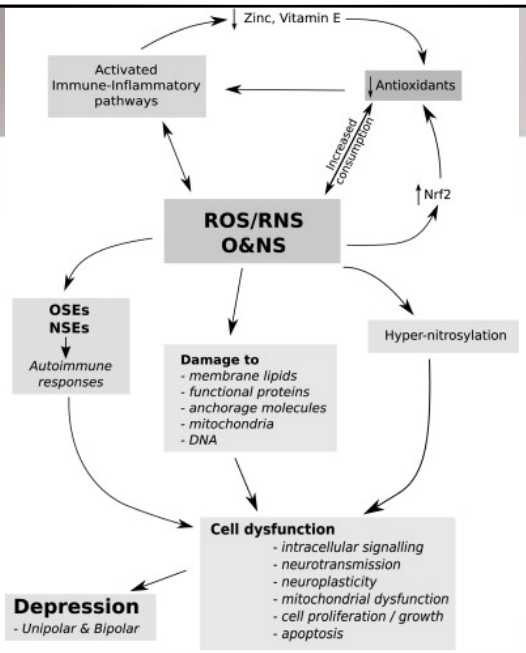
Tangney CC, Tang Y, Evans DA et al. *Neurology* 2009; **72**(4): 361-367

Chicago Health and Ageing Project

- MMA level ⇒ reduced brain volume
- Homocysteine level ⇒ increased white matter lesions and infarcts

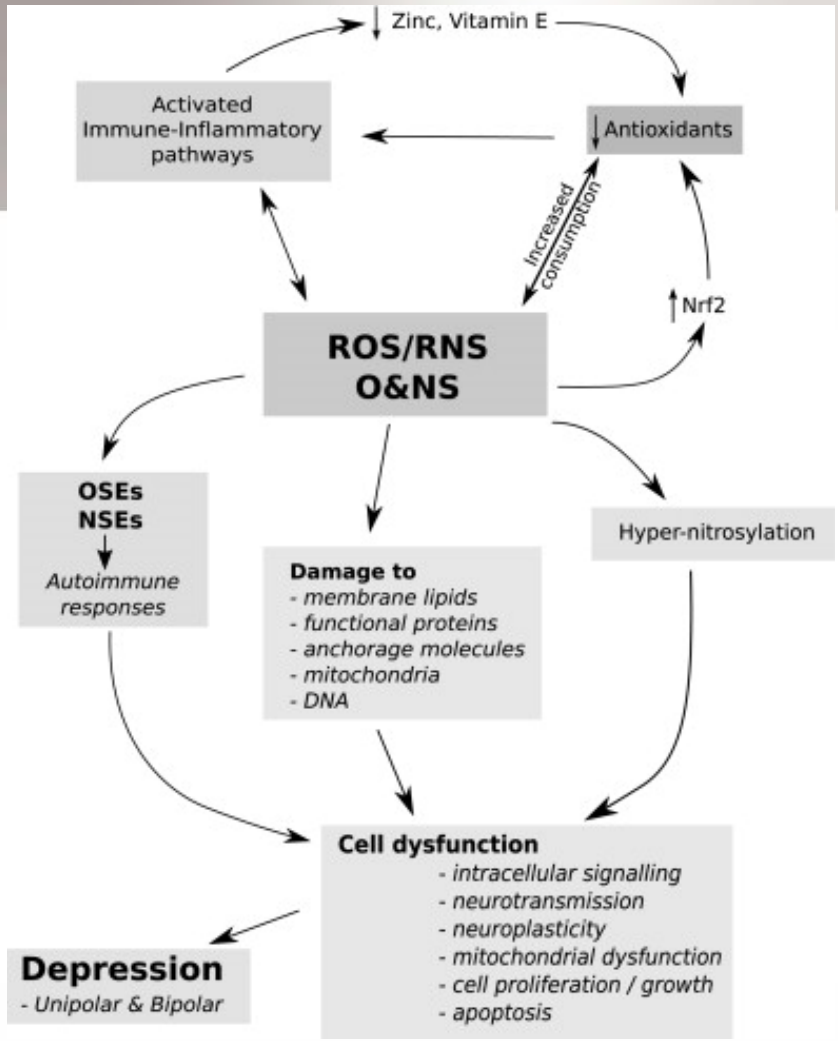
Tangney CC, Aggarwal NT, Li H et al. *Neurology* 2011; **77**(13): 1276-1282

Depression is NI



Moylan S, Berk M, Dean OM et al. *Neurosci Biobehav Rev* 2014; **45**: 46-62

Depression is NI



Moylan S, Berk M, Dean OM et al. *Neurosci Biobehav Rev* 2014; **45**: 46-62

Depression: Key Herbs and Nutrients

- Gold standard evidence for St John's Wort
- Other key herbs: Saffron, Rhodiola and Lavender
- ALC and NAC are important
- Mg and Zn are key elements

Bone KM, Mills SY. *Principles and Practice of Phytotherapy: Modern Herbal Medicine.* 2nd Edition, Elsevier, UK, 2013

Depression: Key Herbs and Nutrients

- B vitamins (especially folate, B12 and B6)
- Omega-3 fatty acids
- Address neuroinflammation drivers, ↑ Nrf2 and additional mitochondrial support as per the case

NAC and Depression

- A recent systematic review:
  - Two trials in depression (one RCT); level B evidence
  - Three trials in bipolar disorder (two RCTs); level A evidence

Deepmala, Slattery J, Kumar N et al. *Neurosci Biobehav Rev* 2015; **55**: 294-321

Bipolar: Key Herbs and Supplements

Intervention	BD (depression)	BD (mania)	Level of evidence: Depression	Level of evidence: Mania
Omega-3	✓		A	D
N-Acetylcysteine	✓		B	C
BCAA		✓	?	B
Inositol	✓		C	?
Choline	✓	✓	C	C
Folic acid	✓		B	C
Magnesium	✓	✓	C	A
Chelated minerals	✓		B	C

Sarris J, Lake J, Hoenders R. *J Altern Complement Med* 2011; **17**(10): 881-890

ADHD: Key Herbs and Supplements

- Ginkgo, Bacopa, Korean Ginseng, Pine Bark (Grape Seed?)
- Zn, Mg and Fe
- Omega-3 fatty acids
- Address neuroinflammation drivers etc

ADHD: 2011 Systematic Review

- Sixteen studies
- Predominant support for Zn, Fe and Pine Bark
- Mixed evidence for omega-3 and ALC

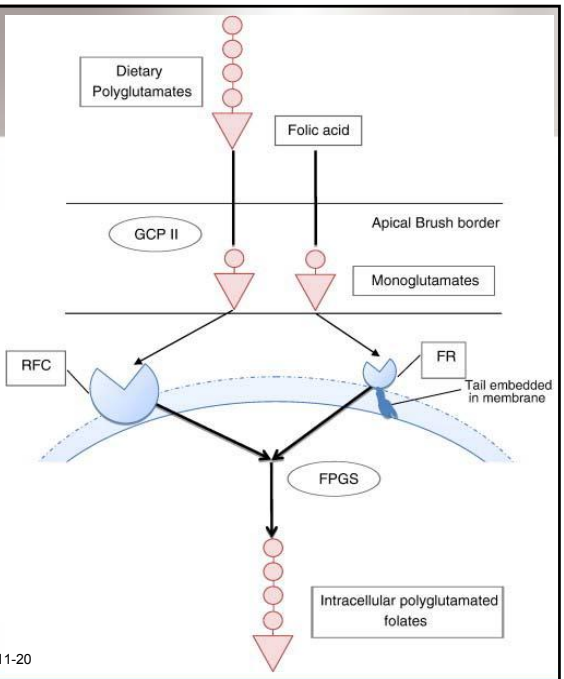
Sarris J, Kean J, Schweitzer I et al. *Complement Ther Med* 2011; **19**(4): 216-227

Mg and ADHD

- Systematic review
- Three trials found, but not RCTs
- Supported use of Mg as effective in ADHD

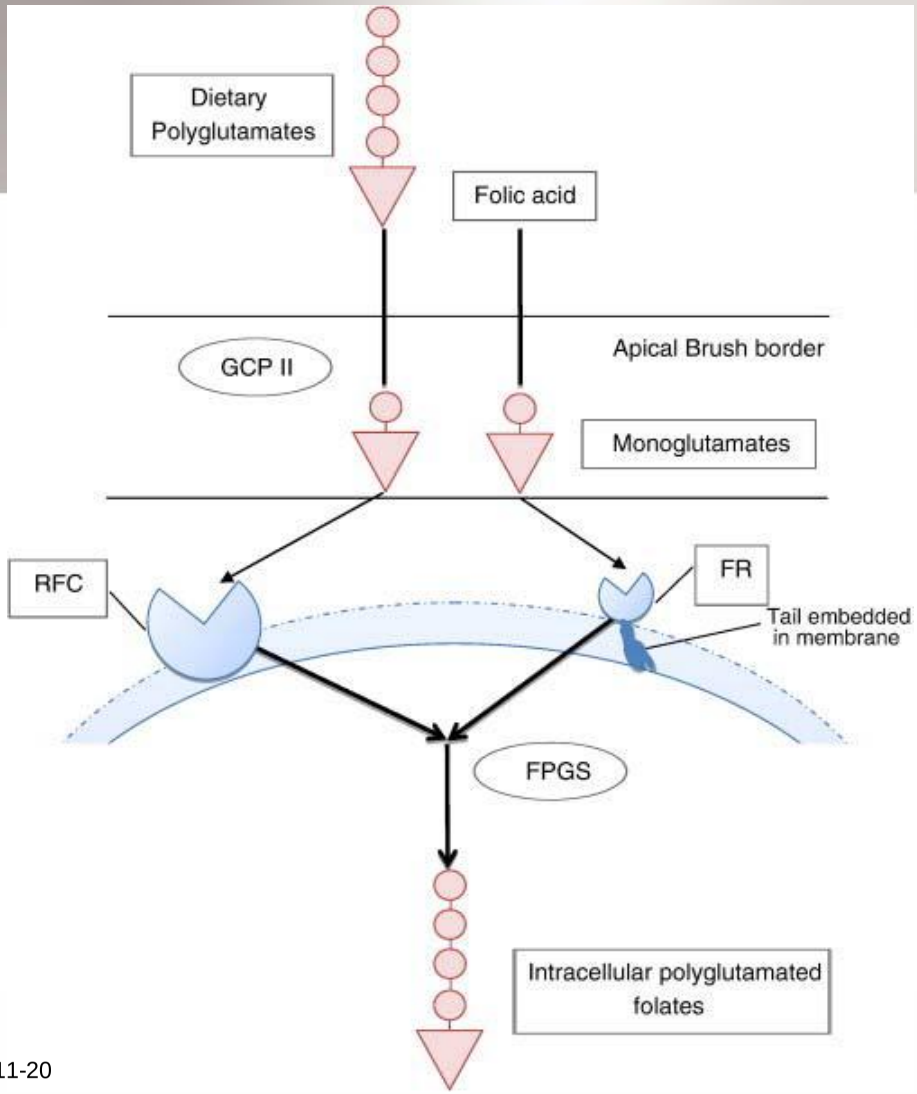
Ghanizadeh A. *Arch Iran Med* 2013; **16**(7): 412-417

Folate Absorption



Nazki FH, Sameer AS, Ganaie BA *Gene* 2014; **533**(1): 11-20

Folate Absorption



Nazki FH, Sameer AS, Ganaie BA *Gene* 2014; **533**(1): 11-20

Cerebral Folate Deficiency Syndrome

- CFDS: newly discovered condition (2002): low folate levels in cerebrospinal fluid (CSF)
- Central nervous system (CNS) deprived of folate
- Caused by FRα AuAbs or genetic defects

Serrano M, Pérez-Dueñas B, Montoya J et al. Drug Discov Today 2012; 17(23-24): 1299-1306

Beyond Infantile-Onset CFDS

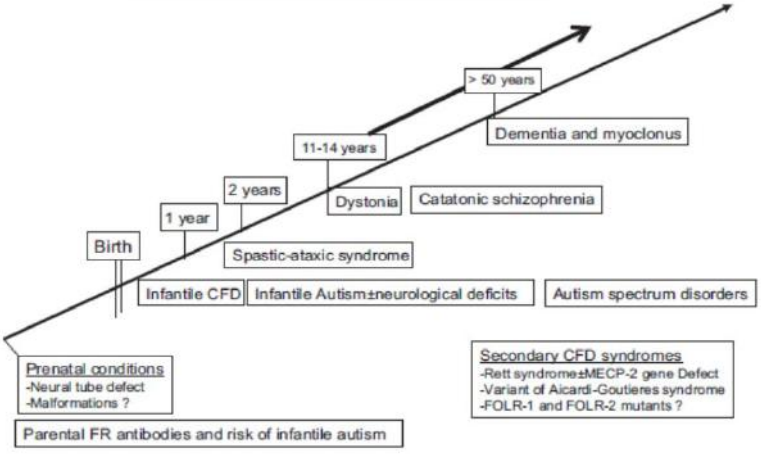


Figure 3 Life tree depicting the recognized CFD syndromes attributed to FR autoimmunity according to age.

Ramaekers V, Sequeira JM, Quadros EV. *Clin Chem Lab Med* 2013; **51**(3): 497-511

Beyond Infantile-Onset CFDS

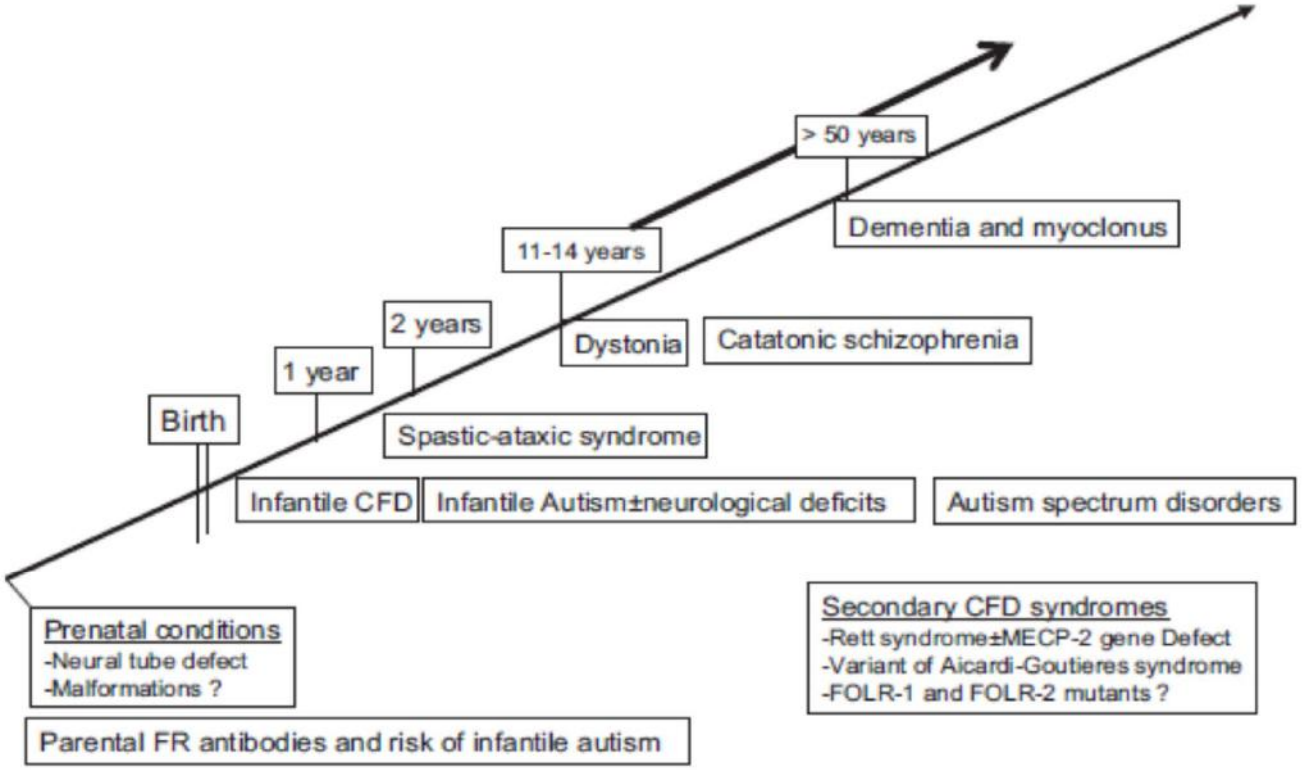


Figure 3 Life tree depicting the recognized CFD syndromes attributed to FR autoimmunity according to age.

Ramaekers V, Sequeira JM, Quadros EV. *Clin Chem Lab Med* 2013; **51**(3): 497-511

CFDS

- Depending on the cause ⇒ a range of severe neurological deficits
- Treated with high dose folate (usually folinic acid, can use 5-methyl-THF)
- Overrides malfunctioning FRα ⇒ uses RFC to access the CSF

Moretti P, Sahoo T, Hyland K et al. *Neurology* 2005; **64**(6): 1088-1090
Hansen FJ, Blau N. *Mol Genet Metab* 2005; **84**(4): 371-373

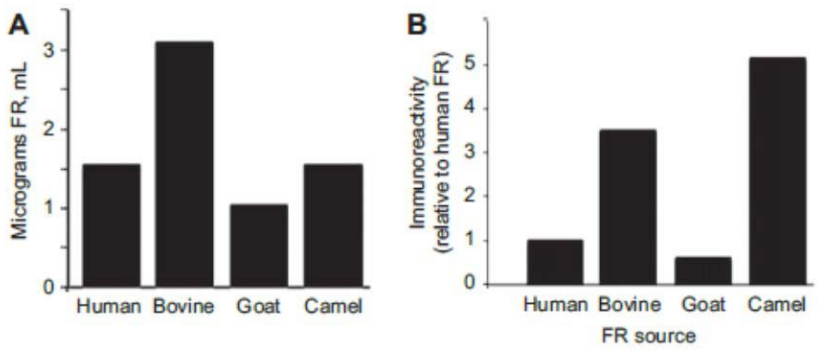
FRα AuAbs and Brain Disorders

Condition	Blocking AuAb	Binding AuAb	Percent prevalence		Reference
			Control		
			Blocking	Binding	
NTD (n=12)	–	75	–	10	8
NTD (n=103)	17	30	13	33	11
CFD (n=28)	89	–	0	–	12
RS (n=33)	24	–	–	–	13
LFA (n=25)	76	–	0	–	14
ASD (n=93)	60	44	–	–	15
ASD (n=75)	46	–	3	–	16

Table 2 Prevalence of FR AuAbs in various conditions.
NTD, mothers with a history of neural tube defect pregnancy; CFD, children with cerebral folate deficiency syndrome; LFA, children with low-functioning autism; ASD, children with autism spectrum disorder; RS, children with Rett syndrome.

Sequeira JM, Ramaekers VT, Quadros EV. *Clin Chem Lab Med* 2013; **51**(3): 545-54

FRα in Milk



Sequeira JM, Ramaekers VT, Quadros EV. *Clin Chem Lab Med* 2013; **51**(3): 545-54

FRα AuAbs and AD?

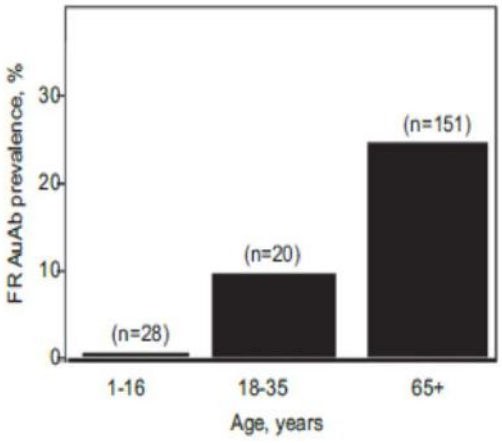


Figure 6 Prevalence of AuAb to FRα with aging.
Serum samples from subjects of different age groups with no known disease condition were analyzed for the presence of blocking AuAb to FRα.

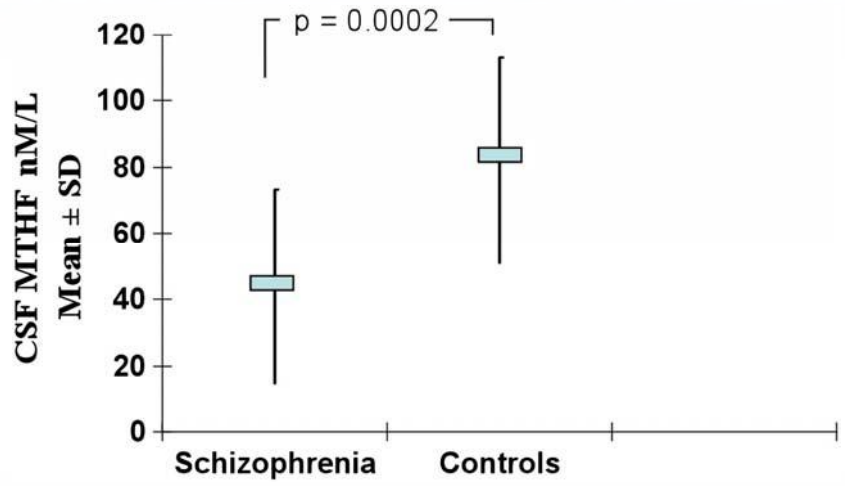
Sequeira JM, Ramaekers VT, Quadros EV. *Clin Chem Lab Med* 2013; **51**(3): 545-554

FRα AuAbs and Schizophrenia

- Schizophrenic patients (n=18) unresponsive to conventional drugs
- 83% positive for FR AuAbs
- Folinic acid to 7 patients resulted in clinical improvement over 6 months

Ramaekers VT, Thöny B, Sequeira JM et al. *Mol Genet Metab* 2014; **113**(4): 307-314

FRα AuAbs and Schizophrenia



Ramaekers VT, Thöny B, Sequeira JM et al. *Mol Genet Metab* 2014; **113**(4): 307-314

Closing Remarks

- Neuroinflammation is a game-changing insight into the best management of a wide range of brain and behavioural disorders
- As well as directing proven herbs and supplements at the condition
- MUST address the drivers of neuroinflammation
- For AD this, and supporting vascular flexibility and microcirculatory function, offer a radical new perspective on prevention and treatment

INTEGRIA HEALTHCARE SYMPOSIUM 2015

Clinical Pearls + Practitioner Perspectives

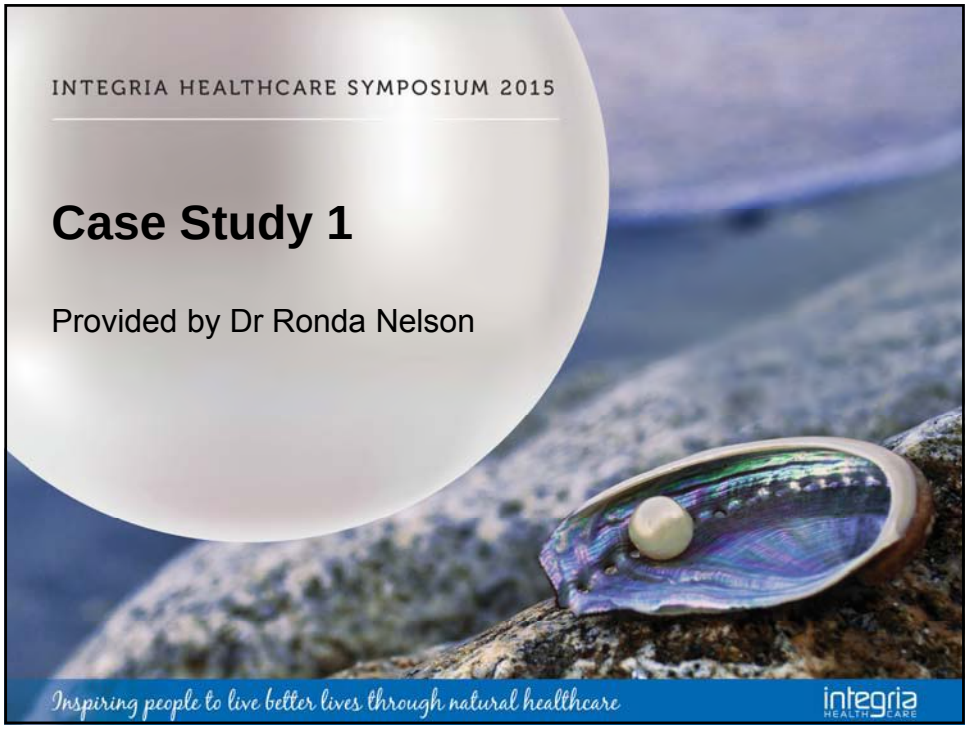
An Experience-Based Approach to Patient Care

24+25 OCTOBER
MELBOURNE

DAY 2

Session 8

Case Study Panel Discussion



Case Study 1: Amy

- Female, 32 years of age
- Height: 168 cm
- Weight: 60 kg
- Primary complaints:
  - Hormones feel “off”
  - History of endometriosis
- History:
  - Menarche at 11 years old
  - Periods were always very painful – learnt how to “tough it out”

Other Presenting Symptoms

- Menstrual cycles becoming irregular – deviating from normal length, now 32-34 days
- Moderate breast tenderness before period
- Heavy flow with some clotting first 2 days
- Acne – moderate

Other Presenting Symptoms

- Mood swings – moderate to severe
- Carbohydrate/sweet cravings
- Moderate backache (mid-back, more on right hand side)
- Abdominal bloating, belching
- Eliminates every 2-3 days

Additional Notes

- Feeds her children better than herself
- Doesn't pay attention to what, when or if she eats
- Doesn't get much sleep – stays up after everyone else goes to bed
- No exercise other than casual walking with friends
- Admits to being an "A-type" personality – always on the go and her mind doesn't stop

Case History

- 20 years old:
  - Periods still very painful
  - Finally went to MD for OCP Rx (getting married) and to ask about pain
  - Some improvement in dysmenorrhoea with OCP
  - Possible kidney/bladder issues investigated, second opinion revealed signs of endometriosis
  - Surgery scheduled

Case History

- 21 years old:
  - Completed first surgery to remove endometrial tissue
  - Grade 4 severe endometriosis
  - Right ovary almost entirely fused to broad ligament
  - Endometrial tissue was everywhere - on both ovaries, uterus, Pouch of Douglas, supporting ligaments
  - Post surgery, MD provided 3.75 mg Lupron SC every month for 6 months to suppress ovarian function
  - Caused severe hot flashes and night sweats
  - Started OCP but side effects were difficult so she discontinued in 6 months (nauseas, bloated, hormonal)
  - After discontinuing, pain returned right away, centered on right side

Case History

- 23 years old
  - Second surgery scheduled with same MD
  - Complete return of endometrial tissue with a few new lesions appearing
  - No post-surgical interventions
  - Completely pain-free for 6 months until the pain slowly started to return

Case History

- 24 years old
  - Laparoscopy performed to check for scar tissue but none found, only returning endometriosis
  - Mid-back pain began returning and symptoms increased
  - Finished college, wanted to conceive but was unsuccessful
  - Diagnostic testing done to determine other comorbidities but none were found

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Case History

- 25 years old
  - Third surgery with a new MD
  - Found endometrial lesions on bladder and intestines, as well as previous locations
  - MD gave Clomid every other cycle and IUI monthly for 6 months
  - Conceived and gave birth as a result
  - No discomfort throughout pregnancy
  - Post-delivery, pain slowly began returning and increased when she finished lactating

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Case History

- 27 years old
  - Fourth surgery with same MD
  - New lesions found on the appendix
  - Started fertility drugs again
  - Successful conception in 2 months
  - Miscarried at 10 weeks
  - Continued 4 additional months of Clomid/IUI (not successful)
  - Pain continued to slowly increase each month

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Case History

- 30 years old
  - Fifth surgery with new MD
  - Abdomen was “almost completely covered” in endometrial lesions
  - MD said she might lose part of her intestines
  - Immediately tried to conceive (without use of Clomid)
  - Successful pregnancy 2 months after surgery

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Case History

- 34 years old
  - Came to my office
  - Past 6 months has felt hormonally "off"
  - Mid-back and right ovarian pain at 6/10 (pre-surgical pain was 10/10)
  - Didn't want to have another surgery but wants to have another child

Initial Recommendations

Herbal tablets providing:	Daily Dose
Silybum marianum (St Mary's Thistle) Extract equivalent to dry seed	8.1 g (providing 96 mg silybin)
Schisandra chinensis (Schisandra) Extract equivalent to dry fruit	4.0 g
Rosmarinus officinalis (Rosemary) Extract equivalent to dry leaf	2.0 g

Herbal tablets providing:	Daily Dose
Echinacea angustifolia and *Echinacea purpurea* root blend	27.6 mg alkylamides

Herbal tablets providing:	Daily Dose
Vitex agnus-castus (Chaste Tree) Extract equivalent to dry fruit	3.0 g

Initial Recommendations (Cont.)

- Chia seeds - 1 tbsp/day
- Support healthy blood sugar by managing diet
- Explore possible sources of exogenous hormone exposure
- Asked her to find out if any family history of endometriosis
- Follow-up in 4 weeks

Week 4 Follow-Up

- No change in menstrual cycle length or flow
- Improvements in:
  - Breast tenderness
  - Elimination (now daily)
  - Mood swings
  - Carbohydrate cravings (now gone)
- Abdominal pain “might” be decreasing slightly
- Energy is better due to dietary changes but still not 100%
- Eliminated two questionable sources of topical hormone exposure
- Mother likely had endometriosis as well

Week 4 Follow-Up Recommendations

Herbal tablets providing:	Daily Dose
Silybum marianum (St Mary's Thistle) Extract equivalent to dry seed	8.1 g (providing 96 mg silybin)
Schisandra chinensis (Schisandra) Extract equivalent to dry fruit	4.0 g
Rosmarinus officinalis (Rosemary) Extract equivalent to dry leaf	2.0 g

Herbal tablets providing:	Daily Dose
Echinacea angustifolia and *Echinacea purpurea* root blend	13.8 mg alkylamides

Herbal tablets providing:	Daily Dose
Vitex agnus-castus (Chaste Tree) Extract equivalent to dry fruit	3.0 g

Week 4 Follow-Up Recommendations

Adrenal Liquid Herbal Blend – 15 mL per day	
Rehmannia 1:2	45 mL
Withania 1:1	30 mL
Echinacea Premium 1:2	20 mL
Licorice High Grade 1:1	5 mL
	100 mL

Week 8 Follow-Up

- Menstrual cycle reduced to 30 days in length
- Bleeding was less – needed fewer pads/tampons
- Mid-back pain definitely decreased
- Discomfort seems to "move" – not in it's usual position
- Overall pain is 4.5/10
- Moods are still stable
- Acne is 75% improved
- Breast tenderness almost completely gone
- Energy is noticeably improved
- Would still like to have one more child, however explained that with higher doses of Chaste tree, this was not likely at this point

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Week 8 Follow-Up Recommendations

Herbal tablets providing:	Daily Dose
Silybum marianum (St Mary's Thistle) Extract equivalent to dry seed	8.1 g (providing 96 mg silybin)
Schisandra chinensis (Schisandra) Extract equivalent to dry fruit	4.0 g
Rosmarinus officinalis (Rosemary) Extract equivalent to dry leaf	2.0 g

Herbal tablets providing:	Daily Dose
Echinacea angustifolia and *Echinacea purpurea* root blend	27.6 mg alkylamides

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Week 8 Follow-Up Recommendations

Adrenal Liquid Herbal Blend – 15 mL per day	
Rehmannia 1:2	45 mL
Withania 1:1	35 mL
Ginger 1:2	15 mL
Licorice High Grade 1:1	10 mL
	100 mL

Female Support Tonic Liquid Herbal Blend – 15 mL per day	
Chaste Tree 1:2	80 mL
Dong Quai 1:2	25 mL
	105 mL

Week 16 Follow-Up

- Energy is exceptional – doesn't ever remember feeling this good
- Diet much better – notices blood sugar disruptions more easily
- Cycle length reduced to 28 days
- Heavy bleeding greatly improved
- Menstrual pain and discomfort continuing to decrease
- Feels like her stomach is flatter
- Thrilled with her progress
- Not loving the liquid herbs but willing to continue
- Wonders why she hadn't explored this avenue before

Week 16 Follow-Up Recommendations

Herbal tablets providing:	Daily Dose
Silybum marianum (St Mary's Thistle) Extract equivalent to dry seed	8.1 g (providing 96 mg silybin)
Schisandra chinensis (Schisandra) Extract equivalent to dry fruit	4.0 g
Rosmarinus officinalis (Rosemary) Extract equivalent to dry leaf	2.0 g

Herbal tablets providing:	Daily Dose
Echinacea angustifolia and *Echinacea purpurea* root blend	27.6 mg alkylamides

Week 8 Follow-Up Recommendations

Adrenal Liquid Herbal Blend – 15 mL per day	
Rehmannia 1:2	45 mL
Withania 1:1	35 mL
Ginger 1:2	15 mL
Licorice High Grade 1:1	10 mL
	100 mL

Female Support Tonic Liquid Herbal Blend – 15 mL per day	
Chaste Tree 1:2	80 mL
Dong Quai 1:2	25 mL
	105 mL

Week 24 Follow-Up

- Broad abdominal pain has resolved
- Only pain remaining is for 2-3 hours on first day of menses
- Centered around navel
- All other presenting symptoms are resolved
- No conception as of yet – wants to continue to try

Week 24 Follow-Up Recommendations

Herbal tablets providing:	Daily Dose
Silybum marianum (St Mary's Thistle) extract equivalent to dry seed	8.1 g (providing 96 mg silybin)
Schisandra chinensis (Schisandra) extract equivalent to dry fruit	4.0 g
Rosmarinus officinalis (Rosemary) extract equivalent to dry leaf	2.0 g

Herbal tablets providing:	Daily Dose
Echinacea angustifolia and *Echinacea purpurea* root blend	13.8 mg alkylamides

Herbal tablets providing:	Daily Dose
Vitex agnus-castus (Chaste Tree) extract equivalent to dry fruit	1.0 g

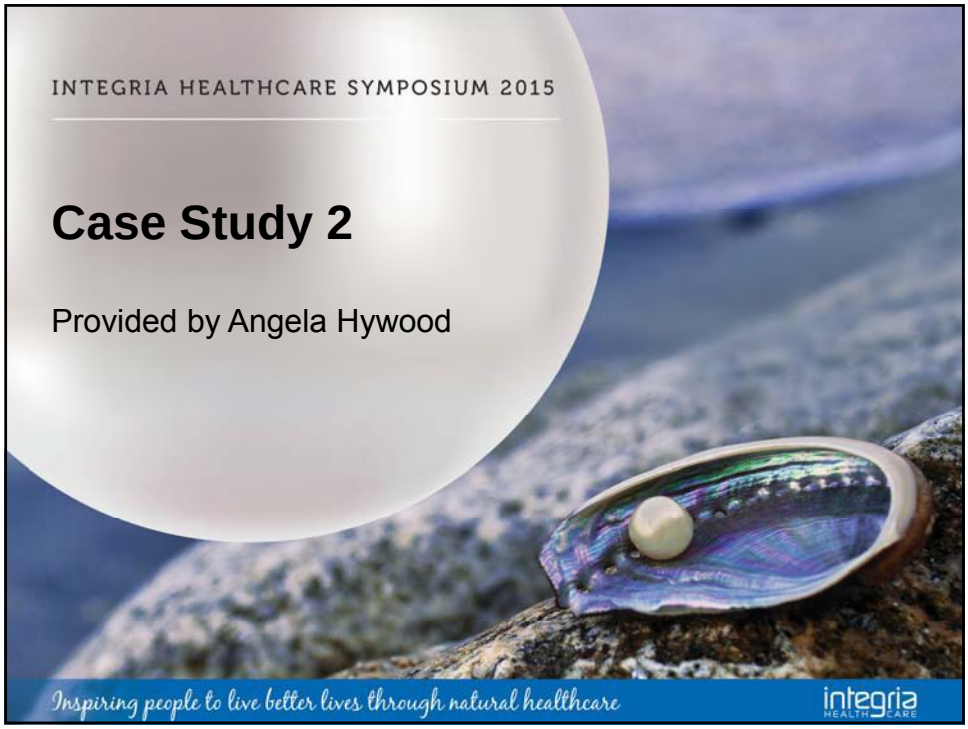
Week 24 Follow-Up Recommendations

Herbal tablets providing:	Daily Dose
Vitis vinifera (Grape Seed) extract equivalent to dry seed	14.4 g (102 mg procyanidins)
Centella asiatica (Gotu Kola) extract equivalent to dry leaf	10.0 g (200 mg triterpene derivatives)
Ginkgo biloba (Ginkgo) extract equivalent to dry leaf	4.0 g (4.8 mg ginkgolides and bilobalide)

General Tonic Liquid Herbal Blend – 15 mL per day	
Rehmannia 1:2	35 mL
Withania 1:1	30 mL
Dong Quai 1:2	30 mL
Licorice High Grade 1:1	5 mL
	100 mL

August 2015 Update

- Patient has been unsuccessful with conception (6 months)
- Pain around navel is yet unresolved
- Lasts 2-3 hours and is quite painful (5/10)
- Some months it is 3/10 but has not fully resolved



Case Study 2: Mrs S

- Female, 36 years of age
- Slim and small framed
  - Height 145cm, weight 42kg, BMI 19 - healthy
- Case History:
  - Overt autoimmune hypothyroidism (Hashimoto's), diagnosed 6 months earlier as post-partum thyroid storm
  - Prescribed 50 mcg Thyroxine per day
  - History of unexplained infertility and 9 x IVF stimulations over for 4 years
  - 2 pregnancies (IVF) 2 healthy children (2.5 years and 6 months)
  - Trouble breastfeeding due to fatigue
  - Baby formula fed from 4 months as breast milk supply diminished

Presenting Symptoms

- Extreme fatigue despite thyroid bloods looking normal and being on medication
- Frequent dizziness
- Poor immunity - recurrent sinus infections
- Gut issues - alternating constipation/loose stools, gassy, bloated
- Feels like she still looks 6 months pregnant

Investigations

	Baseline	Typical Reference Range*
Iron (µmol/L)	12.3	10-30
Transferrin (g/L)	3	1.7-3.0
Total iron binding capacity (TIBC) (µmol/L)	66	45-80
Saturation (%)	20	15-45
Ferritin (µg/L)	56	15-200

- FBC all normal
- Biochemistry all normal

*https://www.rcpa.edu.au/Library/Practising-Pathology/RCPA-Manual/Items/Pathology-Tests

Investigations

Thyroid Function

TSH	* 4.98	mIU/L	(0.40-3.50)
Free T4	10.5	pmol/L	(9.0-19.0)
Free T3	3.7	pmol/L	(2.6-6.0)

Comment
Elevated TSH. If thyroid replacement dosage has not changed recently and compliance is assured, the raised TSH level suggests that increased dosage may be indicated.

Thyroid Antibodies
(Immulite Methods)

Thyroglobulin Ab	<20	IU/mL	(0-40)
Thyroid Peroxidase Ab	* 622	IU/mL	(0-35)

Comment
High levels of thyroglobulin and thyroid peroxidase antibodies are characteristic of Hashimoto's disease. Lower levels can occur in clinically normal persons and with other thyroid disease.

 integria 33

Investigations

Faeces Examination

Specimen	1
Collection date	07-Jan-2015
Appearance	Semi formed
Microscopy Concentrate	No ova, cysts or parasites seen.
Faecal Antigens	
Cryptosporidium Ag	Not Detected
Giardia Ag	Not Detected
Culture	No Salmonella, Shigella or Campylobacter isolated.

 integria 34

Investigations

Random Urine Iodine

U-Creatinine	1.4	mmol/L
Urine iodine	48	ug/L

Comment
WHO classification of iodine deficiency:Urine Iodine levels

Not Iodine deficient:	>100 ug/L urine
Mild Iodine deficiency:	50 - 100 ug/L urine
Moderate Iodine deficiency:	20 - 49 ug/L urine
Severe Iodine deficiency:	<20 ug/L urine

To convert Iodine ug/L to Iodine nmol/L

ug/L x 7.88 = nmol/L

Investigations

Faecal pathogen PCR

Specimen Type	Faeces
Bacteria:	
Campylobacter species	Not Detected
Salmonella species	Not Detected
Shigella species	Not Detected
Yersinia enterocolitica	Not Detected
Aeromonas species	Not Detected
Parasites:	
Giardia lamblia	Not Detected
Cryptosporidium species	Not Detected
Dientamoeba fragilis	Not Detected
Entamoeba histolytica	Not Detected
Blastocystis species	Not Detected

Faeces Examination

Specimen	2
Collection date	07-Jan-2015
Appearance	Formed
Concentrate	No ova, cysts or parasites seen.

Investigations

25 - OH Vitamin D

Vitamin D	74	nmol/L	(51-140)

Comment
According to the Position Statement 'Vitamin D and health in adults in Australia and New Zealand' MJA, 196(11):686-687, 2012, Vitamin D status is defined as:

Mild Deficiency	30 - 49 nmol/L
Moderate Deficiency	12.5 - 29 nmol/L
Severe Deficiency	<12.5 nmol/L

Cortisol

Cortisol am	337	nmol/L	(138-650)
Collection Time	0850	Hrs	

Investigations

Heavy Metals

Serum Copper	13	umol/L	(12-22)
Blood Lead	0.05	umol/L	
Blood Lead	1.0	ug/dL	

Comment
In the absence of occupational exposure, NHMRC recommendation is for blood lead level to be less than 0.48 umol/L (10.0 ug/dL).
With occupational exposure, if the blood lead level exceeds 2.40 umol/L (50.0 ug/dL), NSW Workcover Code of Practice recommendation is removal from further exposure until level falls below 1.90 çmol/L (40.0 ug/dL).

Investigations

HLA DR/DQ Genotyping for Coeliac Disease

Specimen type	EDTA blood
Method	Detection of sequence-specific oligonucleotides (Gen-Probe).
HLA-DR	3, 4
DRB1	03, 04
HLA-DQ	2, 8
HLA-DQA1	03, 05
HLA-DQB1	02, 0302
Interpretation	Genotype susceptible for coeliac disease detected. Heterozygous for the DRB1*03-DQA1*05-DQB1*02 (DQ2) and DRB1*04-DQA1*03-DQB1*03 (DQ8) haplotypes. The presence of the DQ2 and DQ8 antigens confers increased susceptibility to coeliac disease for patient.

Initial Recommendations	Daily Dose	Rationale
Rehmannia glutinosa (Rehmannia) Tyrosine Iodine	3 tabs	Foundational formula for hypothyroidism and adrenal support
Rehmannia glutinosa (Rehmannia) *Bupleurum falcatum* (Bupleurum) *Hemidesmus indicus* (Hemidesmus)	3 tabs	Autoimmune management
Selenium (as selenomethionine) 150 mcg	1 tab	Top up selenium levels due to thyroid antibodies Support thyroxine synthesis
Zinc amino acid chelate Zinc ascorbate Vitamin C	1 tab	Support thyroxine synthesis Support immune function
Glycyrrhiza glabra (GutGard®) *Brassica oleracea var. italica* sprout (BroccoPhane®) Glutamine Vitamin A	1 tsp BID	Gut repair post long-term gluten exposure (gene positive, linked to Hashimoto's) Support Fe absorption
Iron (as ferrous bisglycinate) Folic acid Cyanocobalamin (vitamin B12) Pyridoxal-5-Phosphate (activated B6)	1 tab	Support optimal iron status (subclinical anaemia) Support immune function
Vitamin D3 1000IU	5 sprays	Support optimal D3 levels (deficient) Support immune function

Initial Recommendations	Daily Dose	Rationale
Rehmannia glutinosa (Rehmannia) Tyrosine Iodine	3 tabs	Foundational formula for hypothyroidism and adrenal support
Rehmannia glutinosa (Rehmannia) *Bupleurum falcatum* (Bupleurum) *Hemidesmus indicus* (Hemidesmus)	3 tabs	Autoimmune management
Selenium (as selenomethionine) 150 mcg	1 tab	Top up selenium levels due to thyroid antibodies Support thyroxine synthesis
Zinc amino acid chelate Zinc ascorbate Vitamin C	1 tab	Support thyroxine synthesis Support immune function
Glycyrrhiza glabra (GutGard®) *Brassica oleracea var. italica* sprout (BroccoPhane®) Glutamine Vitamin A	1 tsp BID	Gut repair post long-term gluten exposure (gene positive, linked to Hashimoto's) Support Fe absorption
Iron (as ferrous bisglycinate) Folic acid Cyanocobalamin (vitamin B12) Pyridoxal-5-Phosphate (activated B6)	1 tab	Support optimal iron status (subclinical anaemia) Support immune function
Vitamin D3 1000IU **SLIDE 40**	5 sprays	Support optimal D3 levels (deficient) Support immune function

Primary Tx Plan

- Recommended to follow a gluten free and casein free diet

6 Week Follow-Up

- Energy improved by ~ 60% - still could be better
- Sleep quality improved ("*Nice surprise*")
- Cognition clearer and mood lighter
- Weight stable
- Dizziness mostly improved, but room for improvement
- Gut feels GREAT! No bloating, flat stomach *("This is the best kept secret for post-baby bodies!")*

6 Week Follow-Up

Thyroid Hormones and Antibodies	Baseline	Week 6	Reference Range
TSH (mIU/L)	4.98	2.91	0.40-3.50
Free T4 (pmol/L)	10.5	13.3	9.0-19.0
Free T3 (pmol/L)	3.7	3.7	2.6-6.0
Thyroglobulin Ab (IU/mL)	<20	Not assessed	0-40
Thyroglobulin Peroxidase Ab (IU/mL)	622	Not assessed	0-35

Vitamin D	Baseline	Week 6	Reference Range
25(OH)D3 (nmol/L)	74	112	51-140

6 Week Follow-Up

Random Urine Iodine	Baseline	Week 6	Reference Range
U-Creatinine (mmol/L)	1.4	2.7	
Urine iodine (µg/L)	48	623	>100

Iron Studies	Baseline	Week 6	Typical Reference Range
Iron (µmol/L)	12.3	21.3	10-30
Transferrin (g/L)	3	5	1.7-3.0
Total iron binding capacity (TIBC) (µmol/L)	66	70	45-80
Saturation (%)	20	28	15-45
Ferritin (µg/L)	56	87	15-200

Initial Recommendations	Daily Dose	Rationale
Rehmannia glutinosa (Rehmannia) Tyrosine Iodine	3 tabs	Foundational formula for hypothyroidism and adrenal support
Rehmannia glutinosa (Rehmannia) *Bupleurum falcatum* (Bupleurum) *Hemidesmus indicus* (Hemidesmus)	3 tabs	Autoimmune management
Selenium (as selenomethionine) 150mcg	1 tab	Top up selenium levels due to thyroid antibodies Support thyroxine synthesis
Zinc amino acid chelate Zinc ascorbate Vitamin C	1 tab	Support thyroxine synthesis Support immune function
Glycyrrhiza glabra (GutGard®) *Brassica oleracea var. italica* sprout (BroccoPhane®) Glutamine Vitamin A	1/2 tsp	Gut repair post long-term gluten exposure (gene positive, linked to Hashimoto's) Support Fe absorption
Iron (as ferrous bisglycinate) Folic acid Cyanocobalamin (vitamin B12) Pyridoxal-5-Phosphate (activated B6)	1 tab	Support optimal iron status (subclinical anaemia) Support immune function
Vitamin D3 1000IU	2 sprays	Support optimal D3 levels (deficient) Support immune function

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12 Week Follow-Up

- Energy improved by ~ 80%. Still could be better
- Sleep quality amazing. Can now get by on 6 hours per night, which is all the baby allows
- Cognition great and mood excellent
- Weight stable
- Dizziness 100% resolved
- Gut – no issues at all. No gas, 2 normal solid formed bowel motions per day, flat stomach
- LOVES diet, will never go back to gluten or diary!

Initial Recommendations	Daily Dose	Rationale
Rehmannia glutinosa (Rehmannia) Tyrosine Iodine	3 tabs	Foundational formula for hypothyroidism and adrenal support
Rehmannia glutinosa (Rehmannia) *Bupleurum falcatum* (Bupleurum) *Hemidesmus indicus* (Hemidesmus)	3 tabs	Autoimmune management
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Iron (as ferrous bisglycinate) Folic acid Cyanocobalamin (vitamin B12) Pyridoxal-5-Phosphate (activated B6)	1 tab	Support optimal iron status (subclinical anaemia) Support immune function
Vitamin D3 1000IU **SLIDE 45**	2 sprays	Support optimal D3 levels (deficient) Support immune function

12 Week Follow-Up

Thyroid Hormones and Antibodies	Baseline	Week 6	Week 12	Reference Range
TSH (mIU/L)	4.98	2.91	1.08	0.40-3.50
Free T4 (pmol/L)	10.5	13.3	14.1	9.0-19.0
Free T3 (pmol/L)	3.7	3.7	4.2	2.6-6.0
Thyroglobulin Ab (IU/mL)	<20			0-40
Thyroglobulin Peroxidase Ab (IU/mL)	622		121	0-35