Session 2: Screening
blood tests part II,
Inflammation
Session Objectives

Screening Blood Tests Part II
• Blood lipids (Cholesterol, triglycerides, LDL and HDL)
• Homocysteine

Inflammation
• C-Reactive Protein
• Erythrocyte Sedimentation Rate (ESR)
Cholesterol Panel
Clinical Presentation

- A client may present with the following likely presenting complaints:
  - Personal concerns for poor dietary concerns
  - Past lifestyle factors increasing risk eg: smoker
  - Familial history of Cardio Vascular Disease (CVD)
  - Concern for stress levels
  - NAFLD
  - Referred by medical practitioner to make changes

(Pagana et al. 2015)
Cholesterol

- Blood cholesterol tests tell how much fat is in your blood.

- A total cholesterol level test measures both your LDL (low-density lipoprotein, or "bad cholesterol") and HDL (high-density lipoprotein, or "good cholesterol") levels in milligrams per deciliter (mg/dL).

- Elevated levels of cholesterol in the blood are indicated as a risk for cardiovascular disease.
Clinical Presentation

- In a patient with high blood cholesterol, LDL cholesterol builds up in the inner walls of the arteries that carry blood to the heart and brain.

- Although many people with high cholesterol levels have no symptoms, this narrowing of the arteries (arteriosclerosis) can cause angina (chest pain), heart attack, and stroke.

- Low blood cholesterol rarely causes symptoms, but it may indicate the presence of another disorder. (Pagana et al. 2015)
Cholesterol

- Cholesterol plays a key role in many cellular processes, and is generated by:
  - cells through de novo biosynthesis
  - acquired from exogenous sources (e.g., diet)

- Cholesterol biosynthesis is a complex, multienzyme-catalyzed pathway involving a series of sequentially acting enzymes. This can be influenced by several factors including genetics.

  (Platt, 2015)
Cholesterol

- Due to its presence in the cell membrane, cholesterol forms the function of:

  - permeability
  - fluidity
  - protection against inflammation
  - protection of the myelin sheath of an axon
  - precursor molecule essential for biosynthesis of hormones (vit D, cortisol, aldosterone and sex hormones)

(Pagana et al. 2015)
Cholesterol

- In the liver Cholesterol is converted to bile and then stored in the Gallbladder; Bile Acids and Bile Salts are essential for the absorption and digestion of fat molecules and fat soluble Vitamins – Vitamin A, D, E and K.

- Some recent researches suggest that Cholesterol may act as an antioxidant but this is yet to be confirmed.

(Pagana et al. 2015)
Cholesterol

There are various types of Lipoproteins present in blood. Blood Lipoproteins are classified according to their densities (amount of lipid present in a lipoprotein determines its density, the amount of lipid is inversely proportional with the density).

According to the decreasing order of density the Lipoproteins are:

- High Density Lipoprotein (HDL)
- Low Density Lipoprotein (LDL)
- Intermediate-density Lipoprotein (IDL)
- Very-low-density Lipoprotein (VLDL) and chylomicrons.

(Pagana et al. 2015)
Cholesterol

The most important Lipoproteins for human are LDL and HDL they play an important role in cholesterol metabolism. The main features of LDL and HDL are:

**LDL (Bad Cholesterol)**

LDL is the carrier of cholesterol from the liver to cells. Excess LDL is oxidized and forms foam cells. These then get trapped in the walls of blood vessels and lead to the formation of Atherosclerotic plaques which are considered the main cause of stroke, heart attack and various other cardiovascular diseases.

(Pagana et al. 2015)
Cholesterol

- **HDL (Good Cholesterol):** The function of HDL is to carry cholesterol back to liver for excretion or to the other tissues for further use like production of hormones. This process is known as Reverse Cholesterol transport. HDL is considered good for health.

- It is important to maintain low level of LDL and high level of HDL in blood.

(Pagana et al. 2015)
Cholesterol

- **Triglycerides:** Although not directly related, cholesterol and triglycerides are both considered as important blood lipids, needed for human metabolism.

- The function of Triglycerides is to transport Adipose fat and Glucose.

- High level of Triglycerides may lead to heart diseases and stroke.

(Pagana et al. 2015)
Cholesterol

- When cholesterol blood levels are elevated, it is called Hypercholesterolemia (dyslipidemia)

- This generally happens due to the imbalance of HDL and LDL ratio.

- Low levels of LDL and high level of HDL are needed to maintain proper cholesterol levels, the reverse leads to Hypercholesterolemia (hypolipidemia)

(Pagana et al. 2015)
Cholesterol

Dyslipidemia (High cholesterol) causes & differential diagnosis

- diet & lifestyle (saturated fat intake, smoking)
- metabolic (overweight, high blood sugar)
- diseases (coronary disease)
- inflammation & infection
- endocrine disturbances (thyroid, menopause, pregnancy)
- medications (birth control pills, beta-blockers, estrogen, corticosteroids, and certain diuretics)
- toxin load

(Pagana et al. 2015)
Cholesterol

- Cholesterol overload in the liver can lead to:
  
  - mitochondrial membrane potential changes
  - damaged hepatocytes
  - liver damage e.g.: Non Alcoholic fatty Liver Disease
  - high levels of oxidative stress
  - upregulation of genes related to glutathione homeostasis

(Domínguez-Pérez, 2018)
Cholesterol

Hypolipidemia (Low blood cholesterol) causes and differential diagnosis

- hypothyroidism
- anemia (a low amount of red blood cells)
- malnutrition, or a lack of food
- liver disease
- malabsorption
- rare genetic conditions
- Tangier disease

(Pagana et al. 2015)
Cholesterol

Immune dysregulation

Immune problems can be both a cause and effect of hypercholesterolaemia. This leads to cholesterol accumulation in macrophages and other immune cells and promotes several responses including:

- inflammatory responses
- augmentation of Toll-like receptor (TLR) signalling
- inflammasome activation
- the production of monocytes, neutrophils

- On a cellular level, activation of TLR signalling leads to further cholesterol accumulation and the amplification of inflammatory responses.

(Castrillo, 2013, Duewell, 2010; Fessler, 2013; Moore, 2013)
Cholesterol

Inflammatory response

Cholesterol accumulation through the promotion of inflammatory responses has various actions including:

- during acute infections, HDL becomes dysfunctional and pro-inflammatory
- potentially worsens diseases associated with chronic metabolic inflammation, including atherosclerosis and obesity.
- the interaction of LDL with macrophages in atherosclerotic plaques leads to an increase in inflammatory gene expression and atherogenesis

(Castrillo, 2013, Duewell, 2010)
### Cholesterol and Lipid Profile

<table>
<thead>
<tr>
<th>Reference Ranges (Adult)</th>
<th>Components: (Adult Values)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>• <strong>Total Cholesterol</strong> - &lt;5.20mmol/L</td>
</tr>
<tr>
<td></td>
<td>• <strong>High Density Lipoprotein (HDL)</strong> – [M] &gt;45mg/dL [F] &gt;55mg/dL</td>
</tr>
<tr>
<td></td>
<td>• <strong>Low Density Lipoprotein (LDL)</strong> - &lt;130mg/dL</td>
</tr>
<tr>
<td></td>
<td>• Very Low Density Lipoprotein (VLDL) – 7-32mg/dL</td>
</tr>
<tr>
<td></td>
<td>• <strong>Triglycerides</strong> – [M] 40-160mg/dL [F] 35-135mg/dL</td>
</tr>
</tbody>
</table>

(Pagana et al. 2015; RCPA manual, 2015)
Australian absolute cardiovascular disease risk calculator is useful for analysing the potential clinical implications (in the context of other health factors) of total cholesterol and HDL cholesterol levels.
# Cholesterol: Interpreting the Results

<table>
<thead>
<tr>
<th>CHOLESTEROL</th>
<th>HDL</th>
<th>LDL/VLDL</th>
<th>TRIGLYCERIDES</th>
</tr>
</thead>
</table>
| **HIGH**    | Familial ↑ Cholesterol<br>Hypothyroidism<br>Diabetes mellitus<br>Poor diet<br>Liver disease<br>Nephrotic syndrome<br>Excessive exercise | Familial ↑ HDL<br>Alcohol<br>Liver disease<br>Cushings syndrome<br>Nephrotic syndrome | Familial ↑ LDL<br>Hypothyroidism<br>Alcohol<br>Liver disease<br>Cushings syndrome<br>Nephrotic syndrome<br>Crohn's disease<br>Chronic renal failure | Familial ↑ Tg<br>Hyperlipidaemia<br>Hypothyroidism<br>Hyperthyroidism<br>Malnutrition<br>Syndrome |}
| **LOW**     | Malabsorption<br>Malnutrition<br>Advanced cancer<br>Hyperthyroidism<br>Anaemia<br>Medication | Metabolic syndrome<br>Familial ↓ HDL<br>Hepatocellular disease<br>Nephrotic syndrome | Hypoproteinemia<br>hyperthyroidism | Malabsorption syndrome<br>Malnutrition<br>hyperthyroidism |

(Pagana et al. 2015; Labtests online Australasia, 2018)
Cholesterol

Cholesterol imbalance may therefore be an indicator of other complex illness’ such as

- endocrine dysfunction
- metabolic disorders
- immune dysregulation
- inflammatory responses
- NAFLD (non alcoholic fatty liver disease)
- unexplained infertility in both males and females
- metabolic disorders
- toxin overload
Cholesterol

Management of Hypercholesterolemia:

- Lifestyle changes like regular exercise, weight loss often leads to the reduction of blood cholesterol level.

- However, these may not be effective if elevation is caused by other factors like immune dysregulation or toxic load.

- When interpreting cholesterol pathology, it is important to look at all other factors to ensure an effective treatment strategy. Immune dysregulation is a good example of this.
Discussion

Consider how you may interpret these results and how may these influence your management of:

- age related degenerative disease
- metabolic disorders

What would be the possible differential diagnostic factors related to a 60yr old post menopausal female with fatigue?

What other clinical presentations would you look for in a patient to determine further investigative testing?
Homocysteine
Clinical Presentation

Homocysteine is a (Hcy) is a non-protein α-amino acid, which plays several important roles in human physiology and in the central nervous system.

- Hyperhomocysteinemia (Hhcy) is considered to be an independent risk factor for numerous pathological conditions and it may be both a cause of the disease or impact the disease.

- Test results provide markers for Hhcy which may indicate vitamin B deficiency, or potential as a neurotoxic agent.

(Pagana et al. 2015)
Homocysteine dysregulation is caused by a combination of genetic, nutritional and hormonal factors and is linked to several body systems including the:

1. Cardiovascular system
2. Neurological system
3. Reproductive system

When a strong familial predisposition or early onset vascular disease is noted, homocysteine testing should be performed to determine if genetic or acquired homocysteine excess exists.

(Pagana et al. 2015)
Homocysteine

Clinical presentation of homocysteine may include a combination of several complex disease states including:

- Genetic history of cardiovascular disease
- Learning difficulties & behavioural changes
- Cognitive decline
- Curious about B12 or folate deficiencies & fatigued
- Concerned about development of psychiatric developments such as anxiety or mood changes

(Pagana et al. 2015)
Homocysteine

- Homocysteine is an important predictor of several complex health conditions in conjunction with a thorough holistic assessment such as:

  - coronary, cerebral & peripheral vascular disease.
  - malnutrition (vitamin B12 or folate deficiency)
  - reproductive disorders
  - degenerative age decline
  - renal impairment (poor excretion)
  - environmental toxin accumulation
  - medication use (oral contraceptives, and tamoxifen)

(Pagana et al. 2015; RCPA manual, 2015)
Homocysteine

- Homocysteine is also linked to neurogenerative diseases. These are diseases of the central nervous system with various aetiology and symptoms such as:
  - dementia
  - alzheimer's disease (AD)
  - parkinson's disease (PD)
  - autism

Hyperhomocysteinemia (Hhcy) is considered to be an independent risk factor for numerous pathological conditions under neurodegenerative diseases.

(Sharma, 2015)
Homocysteine

- Hyperhomocysteinemia (Hhcy) is considered to be an independent risk factor for numerous pathological conditions under neurodegenerative diseases.

- Along with genetic factors that are the prime cause of homocysteine (Hcy) imbalance, the nutritional and hormonal factors are also contributing to high Hcy levels in the body.

- Numerous clinical and epidemiological data confirm the direct correlation of Hcy levels in the body and generation of different types of central nervous system disorders, cardiovascular diseases, cancer and others. (Sharma, 2015)
Homocysteine: Interpreting the Results

| HIGH          | Commonly associated with cardiovascular, cerebrovascular and peripheral vascular disease due to the effect of homocysteine to increase vascular endothelial inflammation and plaque formation
|               | B12 deficiency
|               | Folate deficiency
|               | Pernicious anaemia
|               | malnutrition
|               | The above are related to the role of B12 and folate as cofactors for homocysteine metabolism |
| LOW           | Downs Syndrome
|               | Hyperthyroidism |

(Pagana et al. 2015; RCPA manual, 2015)
<table>
<thead>
<tr>
<th>Reference Ranges (Adult)</th>
<th>Components: (Adult Values)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>• Homocysteine – 4-14µmol/L</td>
</tr>
</tbody>
</table>

(Pagana et al. 2015; RCPA manual, 2015)
Homocysteine

Homocysteine imbalance may therefore be an indicator of several complex health conditions.
Consider how you may interpret these results and how may these influence your management of:

- degenerative aging diseases
- female & male health
- metabolic disorders
- neurological disorders

What other clinical presentations would you look for in a patient to determine further investigative testing?
Inflammation: C-Reactive Protein
Clinical Presentation

- A client may present with the following likely presenting complaints:
  - Pain and inflammation in a specific joint
  - Fatigue
  - Concerns for age related decline
  - Concern for infection
  - Generally feeling unwell with low vitality

Pagana et al. 2015
What Is C-Reactive Protein (CRP)

- CRP is a non-specific acute-phase reactant protein that indicates:
  - Inflammatory illness
  - Chronic inflammation
  - Bacterial infectious disease
  - Cardiovascular Disease

Pagana et al. 2015
Testing For CRP

- The CRP test is an extremely nonspecific test. CRP levels can be elevated in any inflammatory condition.

- **C-reactive protein (CRP)** is a blood test marker for inflammation in the body.

- CRP is produced in the liver and its level is measured by testing the blood.

- CRP is classified as an acute phase reactant, which means that its levels will rise in response to inflammation.

(Pagana et al. 2015)
C-Reactive Protein Testing

• CRP interacts with the complement system and serves as an opsonin (enhances phagocytosis) for some microorganisms

• Elevated CRP is linked with increased cardiovascular morbidity and mortality in patients with coronary artery disease.

• Elevated when there is tissue necrosis, malignancies and autoimmune disorders.

• Independent marker for assessing likelihood of recurrent events, including death and myocardial Infarction

(Pagana et al. 2015)
# C-Reactive Protein (CRP)

**C-Reactive Protein (CRP)**

<table>
<thead>
<tr>
<th>Reference Ranges (Adult)</th>
<th>CRP</th>
<th>&lt; 1.0 mg/dL = normal</th>
</tr>
</thead>
<tbody>
<tr>
<td>hsCRP</td>
<td>&lt; 1.0 mg/dL = low risk</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1.0 – 3.0 mg/dL = average risk</td>
<td></td>
</tr>
<tr>
<td></td>
<td>&gt; 3.0 mg/dL = high risk</td>
<td></td>
</tr>
</tbody>
</table>

(CRPs: Non-specific acute-phase reactant protein used to diagnose bacterial infections and inflammatory disorders with associated tissue necrosis. High Sensitivity CRP: measures lower CRP levels associates to estimated cardiovascular risk. (Pagana et al. 2015; RCPA manual, 2015))
CRP/hs-CRP: Risk Factors

CRP levels showing cardiac risk and diseases

[Diagram showing CRP levels and risk factors]

CRP/hs-CRP: Interfering/Risk Factors

- Medications that may increase test results include oestrogen and progesterone.
- Medications that may decrease test results include fibrates, niacin and statins. Both aspirin and statins may help to reduce the inflammation linked to the atherosclerotic process.
- Other drugs, such as non-steroidal-anti-inflammatory drugs (for example ibuprofen) and glucocorticoid drugs, may also lower CRP levels.

(Pagana et al. 2015; RCPA manual, 2015)
## CRP: Interpreting the Results

<table>
<thead>
<tr>
<th>CRP</th>
<th>hsCRP</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Decreased test results</strong></td>
<td>Moderate alcohol consumption, weight loss, increased activity or endurance exercise.</td>
</tr>
<tr>
<td><strong>Elevated test results</strong></td>
<td>Hypertension, elevated BMI, metabolic syndrome, diabetes mellitus, chronic infection, chronic inflammation, low high-density lipoprotein (HDL) and/or triglycerides</td>
</tr>
<tr>
<td>Elevated above 1.0mg/dL</td>
<td>Acute non-infectious inflammatory reactions</td>
</tr>
<tr>
<td></td>
<td>Collagen vascular diseases</td>
</tr>
<tr>
<td></td>
<td>Tissue infarction or damage</td>
</tr>
<tr>
<td></td>
<td>Bacterial infections</td>
</tr>
<tr>
<td></td>
<td>Malignant disease</td>
</tr>
</tbody>
</table>

(Pagana et al. 2015; RCPA manual, 2015)
Inflammation: Erythrocyte Sedimentation Rate
Clinical Presentation

- A client may present with the following likely presenting complaints:
  - Pain, inflammation and joint stiffness
  - Nerve pain
  - Headaches, neck and shoulder pain
  - Concerns for genetic history of Polymyalgia rheumatica, or rheumatoid arthritis
  - Pelvic pain
  - Anaemia, poor appetite, unexplained weight loss,
Erythrocyte Sedimentation Rate (ESR)

- The ESR measure in millimeters how fast the RBCs settle over the period of 1 hour.
- In plasma RBCs usually settle slowly (<15mm/hr). However in the presence of inflammation, infection or necrosis there is an increase in plasma proteins (e.g. Fibrinogen) which cause the RBCs to aggregate (Rouleaux) and settle more rapidly.
- Pregnancy (2\textsuperscript{nd}-3\textsuperscript{rd} Trimester), menstruation, OCP, Vitamin A and some medication can cause elevated levels while anti-inflammatory medications can result in lower ESR levels.

(Pagana et al. 2015; RCPA manual, 2015)
## Erythrocyte Sedimentation Rate (ESR)

### Serum

**ESR:** A measurement of the rate in which RBCs settle in plasma over the period of 1 hour. The ESR is a non-specific test used to detect illnesses associated with infection, inflammation, neoplasm and necrosis.

### Reference Ranges (Adult)

**Components: (Adult Values)**
- **ESR**
  - Male – up to 15mm/hr = normal
  - Female – up to 20mm/hr = normal

*(Pagana et al. 2015; RCPA manual, 2015)*
# Erythrocyte Sedimentation Rate

<table>
<thead>
<tr>
<th>ESR</th>
<th>Conditions</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIGH</td>
<td>Chronic renal failure</td>
</tr>
<tr>
<td></td>
<td>Malignant disease</td>
</tr>
<tr>
<td></td>
<td>Bacterial infection</td>
</tr>
<tr>
<td></td>
<td>Inflammatory disease</td>
</tr>
<tr>
<td></td>
<td>Necrotic disease</td>
</tr>
<tr>
<td></td>
<td>Increased serum proteins</td>
</tr>
<tr>
<td></td>
<td>Severe anaemia</td>
</tr>
<tr>
<td>LOW</td>
<td>Sickle cell anaemia</td>
</tr>
<tr>
<td>(False decrease)</td>
<td>Spherocytosis</td>
</tr>
<tr>
<td></td>
<td>Polycythemia vera</td>
</tr>
<tr>
<td></td>
<td>Low serum fibrin levels</td>
</tr>
</tbody>
</table>

(Pagana et al. 2015; RCPA manual, 2015)
Inflammation: CRP & ESR
CRP & ESR

- ESR is a common initial test when inflammation is being explored due to the cost & complexity of the CRP test.
- Both are markers of inflammation:
  - ESR is a non-specific marker for inflammation whereas CRP is a sensitive indicator of acute phase responses & returns to normal levels quickly with improvement of disease resolution.
  - CRP is not affected by as many other factors as is ESR, making it a better marker of some types of inflammation

(RCPA Manual, 2015)
Consider how you may interpret these results and how may these influence your management of:

- degenerative aging diseases
- female & male health
- metabolic disorders
- paediatric health
- neurological disorders
- cardiovascular disease & cholesterol

What other clinical presentations would you look for in a patient to determine further investigative testing?
References

Castrillo A, et al. Crosstalk between LXR and toll-like receptor signaling mediates bacterial and viral antagonism of cholesterol metabolism. Mol. Cell. 2003;12:805–816. [PubMed] In this paper, activation of TLRs was shown to suppress LXR-responsive genes, notably those involved in cholesterol efflux from macrophages, providing a molecular mechanism to link the response to infectious organisms to suppression of the RCT pathway.


Feingold KR, Grunfeld C. The acute phase response inhibits reverse cholesterol transport. J. Lipid Res. 2010;51:682–684. [PubMed] This paper is an overview of RCT and the acute phase response showing that the acute phase response suppresses RCT at multiple steps.

References


Disorders of cholesterol metabolism and their unanticipated convergent mechanisms of disease.


References


COMMONWEALTH OF AUSTRALIA

Copyright Regulations 1969

WARNING

This material has been reproduced and communicated to you by or on behalf of the Australian College of Natural Medicine Pty Ltd (ACNM) trading as Endeavour College of Natural Health, FIAFitnation, College of Natural Beauty, Wellnation - Pursuant Part VB of the Copyright Act 1968 (the Act).

The material in this communication may be subject to copyright under the Act. Any further reproduction or communication of this material by you may be the subject of copyright protection under the Act.

Do not remove this notice.