

Vitamins

Specialist Portfolio Talk 5th September 2017
Nicola Barlow

Overview

- Vitamins in general
 - Different Types
 - Clinical Effect of Deficiency or Excess
 - DRVs
- Measurement of vitamins

Vitamins

- Organic micronutrients
- Disparate group of organic compounds essential for normal metabolism.
- Cannot be synthesised so must be obtained from the diet.
- Two groups
 - Water soluble
 - Fat soluble

Water Soluble Vitamins

- B Vitamins
 - B₁ = Thiamine
 - B₂ = Riboflavin
 - B₆ = Pyridoxine, pyridoxal and pyridoxamine
 - B₇ = Biotin
 - B₉ = Folic Acid
 - B₁₂ = Cobalamin
 - B₃ = Niacin (nicotinic acid/nicotinamide)
 - B₅ = Pantothenic acid
- Vitamin C = Ascorbic acid

Fat Soluble Vitamins

- Vitamin A = Retinol
- Vitamin D = Ergocalciferol (vitamin D₂) and Cholecalciferol (vitamin D₃)
- Vitamin E = Tocopherols and Tocotrienols
- Vitamin K
 - K₁ = Phylloquinone, Phytomenadione
 - K₂ = Menaquinone
 - K₃ = Menadione

Vitamin A

- Three major functions
 - Present in rods and cones of the retina.
 - Cofactor for making glycoproteins
 - Required for normal epithelial cell function and bone growth.
- Stored in the liver – transported to liver in chylomicrons and away via retinol-binding protein or prealbumin.
- Food sources include liver, fish, egg and milk.

Carotenes

- Retinol can be made from some carotenoids, most importantly from β -carotene (provides ~1/4 of dietary intake).
- Carotenoids are found in plant products, particularly carrots and green veg.
- B-carotene \rightarrow Retinal \rightarrow Retinol

Vitamin A Deficiency

- Deficiency
 - Defective night vision
 - Xerophthalmia leading to keratomalacia and blindness
 - Major cause of blindness in the world.
 - Poor growth of bones and teeth.
 - Increased susceptibility to infections.

Vitamin A Excess

- Excess
 - Toxic in excess.
 - Coarsening and loss of hair.
 - Skin problems
 - Bone fragility
 - Teratogenic (not β -carotene).

B Vitamins

- Isolated deficiencies of B Vitamins in developed countries uncommon (except for folate and B₁₂)
- Generally non-toxic in excess – readily excreted in urine as water soluble – except pyridoxine
- Only limited body storage except B₁₂
- Present in wide variety of animal and plant based foods except B₁₂ (only found in animals)

Vitamin B₁ - thiamine

- Coenzyme, intermediary metabolism especially carbohydrates.
- Found in most foodstuff, especially wheat germ, oatmeal and yeast
- Deficiency – problem in chronic alcoholics, malnutrition 2° to inadequate food intake. Refeeding syndrome
 - Beri-beri – peripheral neuropathy (dry Beri-beri), chronic deficiency, cardiac failure leads to oedema (wet Beri-beri)
 - Wernicke-Korsakoff syndrome – severe, acute deficiency = medical emergency. Confusion, loss of recent memory, peripheral neuropathy
- Only ~30x daily requirement stored in body

Vitamin B₂ - Riboflavin

- Prosthetic groups for numerous metabolic pathways
- Found in milk and offal
- Deficiency
 - No specific deficiency syndrome has been described
 - When it does occur often in conjunction with other nutrient deficiencies.
 - Angular stomatitis
 - Red inflamed tongue
 - Corneal vascularisation and lens opacity

Niacin

- Present as nicotinamide adenine dinucleotide (NAD) and NADP
- Proton acceptor and donor in numerous metabolic reactions.
- Plentiful in animal and plant foodstuffs. Biologically unavailable in some plant foods, e.g., maize
- Deficiency
 - Pellagra – glossitis, dermatitis, diarrhoea and dementia
 - Can be seen as part of carcinoid syndrome.

Vitamin B₆ - Pyridoxine

- Coenzyme for more than 60 enzymes, especially involved in amino acid metabolism
- Widely distributed in foods (some veg sources may be biologically unavailable)
- Deficiency – very rare
 - Glossitis
 - Polyneuropathy
 - Sideroblastic anaemia seen in chronic alcoholics
 - Drugs e.g. isoniazid and penicillamine

Vitamin B₇ - Biotin

- Prosthetic group for carboxylase enzymes
- Widely distributed in foods especially offal, milk and eggs
- Deficiency – very rare
 - Dermatitis, hair loss
 - Problem if raw eggs are eaten – they contain avidin that binds biotin with high affinity

Pantothenic acid

- Present in bodies as coenzyme A
- Essential for metabolism of carbohydrates, fats and proteins.
- Major component of the Krebs cycle
- “Available everywhere”. Rich sources include liver, meat, cereals, milk, egg yolk and fresh veg
- Deficiency
 - No specific naturally occurring syndrome

Vitamin B₉ – Folic acid

- Major active form is tetrahydromethylfolate (THF)
- Used for single carbon transfers in multiple metabolic reactions and essential for DNA synthesis
- Found in liver, legumes, nuts, yeast extract, wholemeal bread
- Deficiency
 - Due to dietary insufficiency, malabsorption, excess utilisation (e.g. pregnancy, lactation) or loss (e.g. dialysis).
 - Drugs e.g. methotrexate
 - Macrocytosis
 - Megaloblastic anaemia

Vitamin B₁₂

- It is a cobalamin – contains cobalt
- Coenzyme – essential for the synthesis of DNA
- Stored in the liver
- Found in animal products (including fish & milk)
- Deficiency
 - Macrocytic, megaloblastic anaemia
 - Neurological disorder
 - Vegans may be at risk
 - Pernicious anaemia and malabsorption major causes

Vitamin C – ascorbic acid

- Reducing agent, acting as an antioxidant and scavenger of free radicals
- Essential for formation of collagen and synthesis of carnitine, serotonin, dopamine and bile salts
- Promotes the absorption of inorganic iron
- Found particularly in citrus fruits, tomatoes, potatoes and leafy vegetables. Cooking markedly reduces vitamin C content

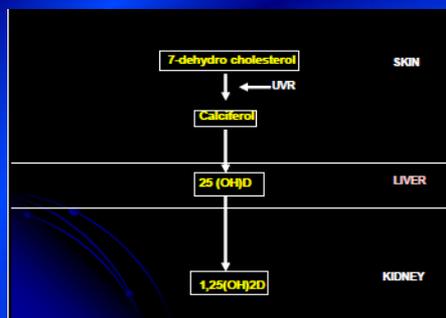
Vitamin C Deficiency

- Scurvy
 - Perifollicular haemorrhages
 - Swollen, bleeding gums
 - Easy bruising
 - Spontaneous haemorrhage
 - Failure of wound healing
 - Anaemia
- Treat by giving ascorbate

Vitamin D

- Ergocalciferol is semi-synthetic, present in food fortified with vitamin D
- Cholecalciferol is produced in the skin by the action of UV radiation
- Also present in dairy products and egg yolk
- Endogenous synthesis is the major source

Production



Vitamin D Function

- Traditionally involved with bone metabolism and calcium homeostasis
- Now thought to have many other roles including cancer, immune modulation, cardiovascular disease, multiple sclerosis and diabetes
- Active hormone is calcitriol
 - Acts on gut and bone with PTH

Vitamin D Deficiency

- Deficiency
 - Rickets in children
 - Osteomalacia in adults
 - Bone pain, tenderness and myopathy
- Risk factors
 - Dietary deficiency (e.g. exclusively breast fed)
 - Low exposure to sunlight
 - Malabsorption
 - Drugs

Vitamin D Deficiency

- Deficiency
 - Calcitriol deficiency seen in chronic renal failure
 - Inherited disorders
 - Vitamin D-dependent rickets Type I – decrease hydroxylation to 1alpha-25-OHD
 - Vitamin D-dependent rickets Type II – defect in the receptor for calcitriol

Vitamin D Excess

- Excess
 - Not possible through excessive exposure to sunlight
 - Calcitriol is tightly regulated
 - However can have excessive 25-OHD which is thought to have some of the biological activity of calcitriol.
 - Supra-physiological supplementation
 - Sarcoidosis
 - Hypercalcaemia leading to metastatic calcification

Vitamin E

- Powerful antioxidant in cell membranes
- Found in vegetable oil and cereals. Supplemented in formula milk
- Deficiency
 - Rare
 - Haemolytic anaemia
 - Thrombocytopaenia
- Nutritional requirement proportional to intake of polyunsaturated fatty acids but foods rich in these contain a lot of vitamin E

Vitamin E

- Excess
 - Predisposition to bleeding (interference with metabolism of vitamin K).
 - However low toxicity generally

Vitamin K

- Essential for blood clotting
- Some vitamin K is synthesised by colonic bacteria
- Widespread availability in vegetables
- Deficiency
 - Rare
 - Bleeding tendency
 - Can occur in fat malabsorption

Vitamin K and newborns

- Healthy newborns are at risk of vitamin K deficiency as a result of poor placental transfer of vitamin K and initial sterility of infants gut
- Breast milk is also a poor source
- Babies are given prophylactic Vitamin K shortly after birth to prevent haemorrhagic disease of the newborn

Nutritional Requirements

- The amount of each nutrient needed by people to stay healthy is called the nutritional requirement.
- These are different for each nutrient and also vary between individuals and life stages, e.g. women of childbearing age need more iron than men.

Nutritional Requirements

- Individual requirements of each nutrient are related to:
 - Age
 - Gender
 - level of physical activity
 - state of health
- Some people absorb or utilise nutrients less efficiently than others and so will have higher than average nutritional requirements.

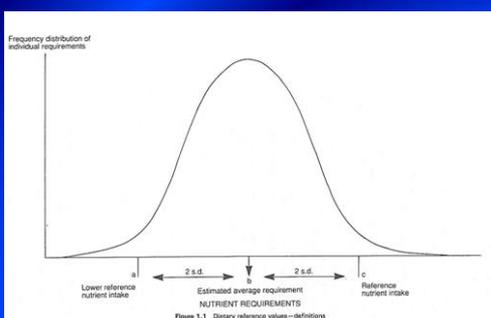
Dietary Reference Values

- COMA (Committee on Medical Aspects of Food and Nutrition Policy)
 - 1991 report – Dietary Reference Values (DRVs) for Food Energy and Nutrients for the UK
- COMA superseded by SACN (Scientific Advisory Committee on Nutrition)
- No real changes since 1991

DRVs

- DRVs are estimates of the requirements for groups of people and are not recommendations or goals for individual people.
- There are three types of estimates:
 - Estimated Average Requirements (EARs)
 - Reference Nutrient Intakes (RNIs)
 - Lower Reference Nutrient Intakes (LRNIs).

DRVs



RNIs for Vitamins

Age	Thiamin mg/d	Riboflavin mg/d	Niacin (niotinic acid equivalent) mg/d	Vitamin B6 mg/d ¹	Vitamin B12 µg/d	Folate µg/d	Vitamin C mg/d	Vitamin A µg/d	Vitamin D µg/d
0-3 months	0.2	0.4	3	0.2	0.3	50	25	350	8.5
4-6 months	0.2	0.4	3	0.2	0.3	80	25	380	8.5
7-9 months	0.2	0.4	4	0.3	0.4	80	25	350	7
10-12 months	0.3	0.4	5	0.4	0.4	50	25	350	7
1-3 years	0.5	0.6	6	0.7	0.5	70	30	400	7
4-6 years	0.7	0.8	11	0.9	0.8	100	30	400	-
7-10 years	0.7	1.0	12	1.0	1.0	150	30	500	-
Males									
11-14 years	0.9	1.2	15	1.2	1.2	200	35	500	-
15-18 year	1.1	1.3	18	1.5	1.5	200	40	700	-
19-50 years	1.0	1.3	17	1.4	1.5	200	40	700	-
50+ years	0.9	1.3	10	1.4	1.5	200	40	700	**
Females									
11-14 years	0.7	1.1	10	1.0	1.2	200	35	600	-
15-18 year	0.8	1.1	14	1.2	1.5	200	40	600	-
19-50 years	0.8	1.1	13	1.2	1.5	200	40	600	-
50+ years	0.8	1.1	12	1.2	1.5	200	40	600	**
Pregnancy	+0.1 ^{***}	+0.3	-	-	-	+100	+10	+10	10
Lactation									
0-4 months	+0.2	+0.5	+2	-	+0.5	+50	+20	+350	10
4-6 months	+0.2	+0.5	+2	-	+0.5	+50	+30	+350	10

¹No increment ^{**}After age 55 the RNI is 10 µg/d for men & women ^{***}For last trimester only [†]Based on protein providing 14.7% of EAR for energy

Measurement of Vitamins

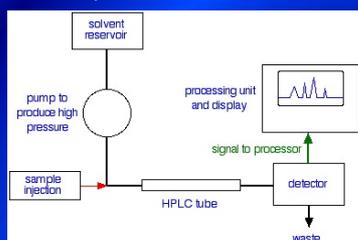
- Most vitamins can be measured
- Most commonly measured are
 - Vitamin A
 - Vitamin D
 - Vitamin E
 - Carotenes
 - B₁₂
 - Folate

Methodologies

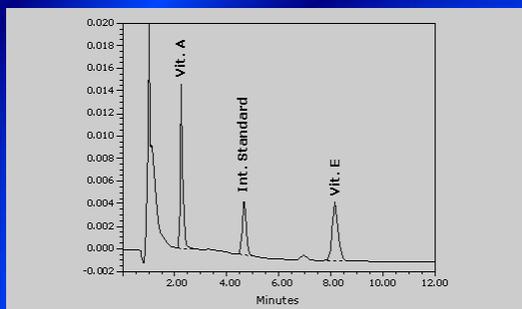
- HPLC
- LC/MS/MS
- Immunoassay
 - RIA
 - EIA

HPLC

- High Performance Liquid Chromatography
 - Mobile Phase and Stationary Phase
 - High Pressure
 - Multiple detector types available



HPLC



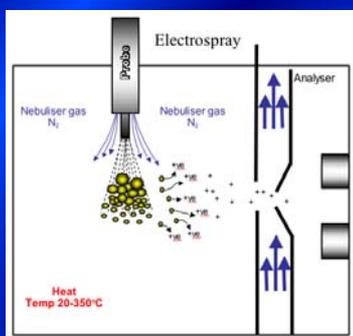
HPLC

Advantages	Disadvantages
Relatively Low Consumable Cost	Relatively High Equipment Cost
Can be automated	Skilled Operators
Multiple Detectors	Manual Extractions
Versatile Equipment	Lack of Specificity (co-eluting peaks)
Stable	Batched workload
Sensitive	Fairly Slow
	Low Throughput

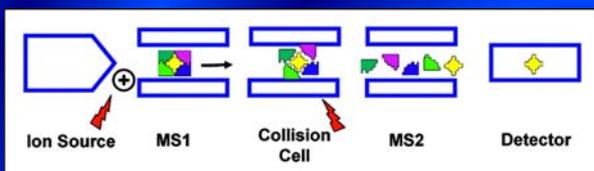
LC/MS/MS

- Liquid Chromatography Tandem Mass Spectrometry
 - HPLC coupled to mass spectrometers
 - 3 MSs – 2 for detecting, one as a collision cell
 - 3 measuring parameters (at least) so often very specific methodology
 - Mass-to-charge ratio (m/z) – need ionised particles
 - Different operational modes

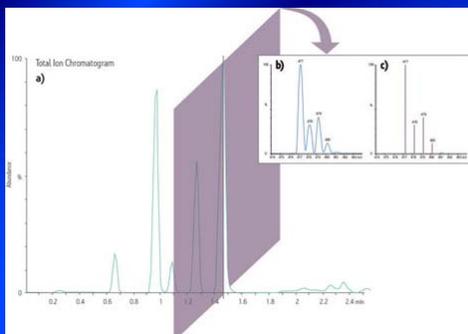
LC/MS/MS

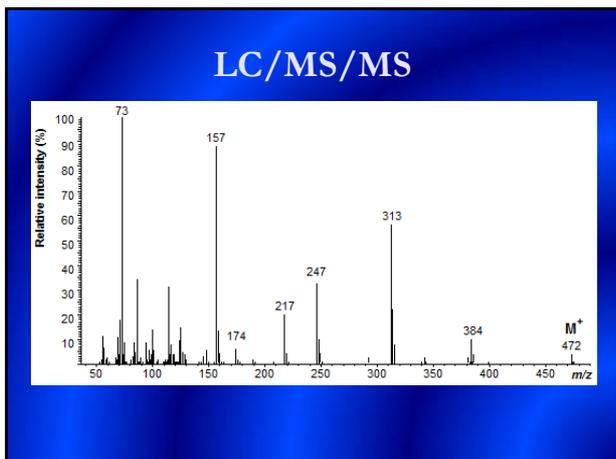


LC/MS/MS



LC/MS/MS





LC/MS/MS

Advantages	Disadvantages
Relatively Low Consumable Cost	High Equipment Cost
Can be automated	Skilled Operators
Versatile Equipment	Manual Extractions
Stable	Batched workload
Sensitive	Fairly Slow
Specific	Low Throughput
Multiple modes	

Immunoassay

Advantages	Disadvantages
Open Access	High Equipment Cost
Quick	Expensive Reagents
Fully Automated Process	Interferences
High Throughput	Lack of Specificity
