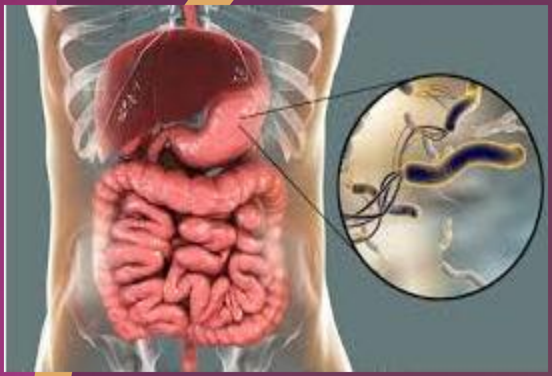
Nutritional Microbiology



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Ministry of Health & Population
وزارة الصحة والسكان

Acknowledgments

This two-year curriculum was developed through a participatory and collaborative approach between the Academic faculty staff affiliated to Egyptian Universities as Alexandria University, Ain Shams University, Cairo University , Mansoura University, Al-Azhar University, Tanta University, Beni Souef University , Port Said University, Suez Canal University and MTI University and the Ministry of Health and Population(General Directorate of Technical Health Education (THE). The design of this course draws on rich discussions through workshops. The outcome of the workshop was course specification with Indented learning outcomes and the course contents, which served as a guide to the initial design.

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توصيف مقرر دراسى

1- بيانات المقرر		
الرمز الكودى :	**اسم المقرر :** **Nutritional microbiology**	**الفرقة /المستوى :** **الثانيه**
التخصص :	**عدد الوحدات الدراسية : 2 نظرى 2عملى**	

2- هدف المقرر: 2-Course Goals (overall aim):	This is a course equip students with knowledge and basic skills related to nutritional microbiology and the relevance of microbes to human disease.
3- المستهدف من تدريس المقرر : 3-Intended Learning Outcomes (ILOs):	
ا. المعلومات والمفاهيم : **A-Knowledge and Understanding:**	**By the end of the course every student will be able to:** A.1. Explain how the microbiome is important for maintaining human health. A.2 Identify the difference between breast milk and formula. A.3. Recognize interaction among gut microbiota, host and food. A.4.Describe the interaction between malnutrition and infection. A.5. Explain how a deficiency, excess or imbalance of nutrients may affect immunity. A.6.Explain how inflammation is involved in chronic disease: obesity, cardiovascular disease and type 2 diabetes. A.7.Describe the role of probiotics and prebiotics in treating various medical conditions. A.8. Identify the principles of food safety.
ب- المهارات الذهنية : **B- Intellectual Skills:**	B.1. Discuss the main dietary goals in the management of IBD. B.2. Discuss the main dietary goals in the management of food allergy. B.3. Analyze a case study problem about infectious diseases. B.4.Discuss the role of vitamins in immune function: A, B-6, B-12, C, D, E, beta-carotene and folic acid. B.5. Discuss the role of minerals in immune function: zinc, copper, iron, magnesium and selenium. B.6.Discuss the concepts of epigenetics and nutrigenomics and how they relate to immunity and diet.
ج- المهارات المهنية الخاصة بالمقرر: **C- Professional and Practical Skills:**	C.1. Implement nutritional management of HIV infection, IBD and food allergy. C.2.Use the appropriate probiotic or prebiotic for various medical conditions. C.3.Apply the reliable, genomic information of nutrigenomics with the goal of improving health through personalized nutrition.

D.1. Work in collaboration as a member of an interdisciplinary team to solve a case about infectious diseases. D.2. Gather, organize and appraise information including the use of information technology where applicable in diagnosis of infectious diseases. D.3. Present the reliable medical information of nutrigenomics, prebiotics and probiotics in written, oral and electronic forms.	د- المهارات العامة : **D-General and Transferable Skills:**
1. Microbiota in health and disease. 2. Gut microbiota. 3. The microbiota in inflammatory bowel disease 4. Food microbiology and food safety 5. Interaction between malnutrition and infection 6. Nutrition and immunity. 7. Food allergy 8. HIV/AIDS 9. Microorganisms in Human Welfare: genetically engineered organisms, probiotics and single cell proteins 10. Prebiotics. 11. Food and water borne diseases. 12. Molecular genetics, epigenetics and nutrigenomics.	**4- محتوى المقرر:** 4-**Course Content**

5.1. Lecture/ week (for two hours each) 5.2. Problem solving 5.3. Small group discussion	**5- أساليب التعليم والتعلم** 5- **Teaching and Learning Methods:**
	6- أساليب التعليم والتعلم للطلاب ذوى القدرات المحدودة 6- **Teaching and Learning Methods for students with limited capabilities:**
7- Student Assessment Methods:	**7- تقويم الطلاب :**
- Written exam. -MCQs exam. - Quizzes, assignments, and practical exam.	**أ- الأساليب المستخدمة** **A-Methods used**
Midterm Exam: At the ---------- week Final Exam: At the end of the course	**ب- التوقيت** **B- Timing of Assessment:**
final written examination:60% 80 marks semester work: 5% quiz 3 marks Attendance : 2 marks Midterm ; 10 marks Assignment: 10 marks **total:100%**	**ج- توزيع الدرجات** **C-Distribution of Scores:**

8- قائمة الكتب الدراسية والمراجع : 8-List of Textbooks and References:

أ- مذكرات **A- Course Notes:**	

ب- كتب ملزمة **B- Essential Books (Textbooks):**	
ج- كتب مقترحة **C- Recommended Books:**	-Microbiology: An Introduction/Media Update, 7th Tortora, Gerard J.; Case, Christine L.; Funke, Berdell R. - Case Files Microbiology, Third Editionby Eugene Toy and Cynthia R. Skinner DeBord -Clinical Case Studies For The Nutrition Care Process by Elizabeth Zorzanello Emery - Nikolaos Katsilambros, Charilaos Dimosthenopoulos, Meropi Kontogianni and Evangelia Manglara(2010) Clinical Nutrition in Practice
د- دوريات علمية أو نشرات ...... الخ **Periodicals, websites,…. etc.**	Google scholar, pubmed search.

Course Contents

Course Description:

This is a course through which the students should gain the knowledge and insight into therapeutic Nutrition and must be able to:

Know latest knowledge and basic skills in the theoretical and practical aspects of microbiology and immunology and the relevance of microbes to Therapeutic Nutrition .

Core Knowledge

By the end of the course every student will be able to:

A.1 Explain how the microbiome is important for maintaining human health.

A.2 Discuss the difference between breast milk and Formula.

A.3. Recognize Interaction among gut microbiota, host and food.

A.4.Describe the interaction between malnutrition and infection.

A.5. Explain how a deficiency, excess or imbalance of nutrients may affect immunity.

A.6.Explain how inflammation is involved in chronic disease: obesity, cardiovascular disease and type 2 diabetes.

A.7.Describe the role of probiotics and prebiotics in treating various medical conditions.

A.8. Understand the principles of food safety.

Core Skills

By the end of this course, students should be able to:

B.1. Discuss the The main dietary goals in the management of IBD.

B.2. Discuss the The main dietary goals in the management of food allergy.

B.3. Analyse a case study problem about infectious diseases.

B.4.Discuss the role of vitamins in immune function: A, B-6, B-12, C, D, E, beta-carotene and folic acid.

B.5. Discuss the role of minerals in immune function: zinc, copper, iron, magnesium and selenium.

B.6.Discuss the concepts of epigenetics and nutrigenomics and how they relate to immunity and diet.

v

Course Overview

		Methods of Teaching/Training with Number of Total Hours per Topic		
ID	**Topics**	**Lecture**	**Assignments**	**Practical**
1	Micro biota in health and disease	**2**		**2**
2	Gut micro biota.	**2**		**2**
3	The micro biota in inflammatory bowel disease	**2**		**2**
4	Food microbiology and food safety	**2**		**2**
5	Interaction between malnutrition and infection	**2**		**2**
6	Nutrition and immunity.	**2**		**2**
7	Food allergy	**2**		**2**
8	HIV/AIDS	**2**		**2**
9	Microorganisms in Human Welfare: genetically engineered organisms, probiotics	**2**		**2**
10	Prebiotic	**2**		**2**
11	Food and water borne diseases	**2**		**2**
12	Molecular genetics, epigenetics and nutrigenomics	**2**		**2**
	Total hours (48)	**24**		**24**

Terminology

Acquired immunodeficiency syndrome (AIDS)is a disorder resulting from the infection with the human immunodeficiency virus (HIV), which leads to a profound immunosuppression and high susceptibility to life-threatening

Allergen: A substance (antigen, see below) that provokes an allergic reaction.

Antibodies: Substances produced by B cells that react with antigens and prepare them for destruction.

Antigen: Asubstance that triggers an immune response.

Antioxidant: Any substance that can delay or inhibit oxidation.

Atherosclerosis: A degenerative disease of arteries in which there is thickening caused by the accumulation of material (plaque) beneath the inner lining, eventually restricting blood flow. The plaque characteristically contains cholesterol and macrophages.

Cloning is the process of creating many identical copies of a sequence of DNA. The target DNA sequence is inserted into a cloning vector

Epigenetics is the study of heritable phenotype changes that do not involve alterations in the DNA sequence. Epigenetics most often denotes changes that affect gene activity and expression, but can also be used to describe any heritable phenotypic change.

Fermentation is one-way microorganisms can change a food. Yeast, especially Saccharomyces cerevisiae**,** is used to leaven bread, brew beer and make wine.

Food allergy refers to specific reactions that result from an abnormal immunological response to a food and which can be severe and life-threatening and triggered by minute amounts of the allergen.

Food microbiology is the study of the microorganisms that inhabit, create, or contaminate food.

Food safety is a major focus of food microbiology. Pathogenic bacteria, viruses and toxins produced by microorganisms are all possible contaminants of food.

Gene amplification is a procedure in which a certain gene or DNA sequence is replicated many times in a process called DNA replication.

Highly active antiretroviral therapy (HAART) is a term used to describe the use of a combination of antiretroviral drugs for the treatment of HIV infection.

Human microbiome refers specifically to the collective genomes of resident microorganisms.

Human Microbiome Project took on the project of sequencing the genome of the human microbiota, focusing particularly on the microbiota that normally inhabit the skin, mouth, nose, digestive tract, and vagina.

Inflammation is a basic process whereby tissues of the body respond to injury.

Malnutrition is a condition that results from eating a diet in which one or more nutrients are either not enough or are too much such that the diet causes health problems. It may involve calories, protein, carbohydrates, vitamins or minerals.

Microbiota is a collective term for the micro-organisms that live in or on the human body. Specific clusters of microbiota are found on the skin or in the gastrointestinal tract, mouth, vagina and eyes.

Molecular genetics is the field of biology that studies the structure and function of genes at a molecular level and thus employs methods of both molecular biology and genetics.

Non-allergic food intolerance refers to reactions to food that can result from a number of causes, none of which is mediated by the immune system (e.g. pharmacological effects, enzyme deficiencies, irritant and toxic effects).

Nutrigenetics: studies the effect of genetic variations on the interaction between diet and health with implications to susceptible subgroups.

Nutrigenomics: studies the effect of nutrients on health through altering genome, proteome, metabolome and the resulting changes in physiology.

Nutritional genomics is a science studying the relationship between human genome, nutrition and health.

opportunistic infections and malignancies

Personalized nutrition uses familial, genetic, or metabolomics information to interpret an individual's health risk profile.

Prebiotics are food ingredients that induce the growth or activity of beneficial microorganisms (bacteria and fungi). The most common example is in the gastrointestinal tract, where prebiotics can alter the composition of organisms in the gut microbiome.

Probiotics are microorganisms that are claimed to provide health benefits when consumed.

Recommended daily amount (RDA)for a nutrient indicates the average daily dietary intake level considered sufficient to meet the requirements of nearly all (97–98%) healthy individuals.

Single-cell protein (SCP) refers to edible unicellular microorganisms. The biomass or protein extract from pure or mixed cultures of algae, yeasts, fungi or bacteria may be used as an ingredient or a substitute for protein-rich foods, and is suitable for human consumption or as animal feeds.

Synbiotics are food ingredients or dietary supplements combining probiotics and prebiotics in a form of synergism.

List of abbreviations

AIDS	acquired immunodeficiency syndrome
ASD	autistic spectrum disorder
B. lactis	Bifidobacterium lactis
BCFA	branched-chain fatty acids
BCM	body cell mass
BHA	butylated hydroxyanisole
BHT	butylated hydroxytoluene
C. botulinum	Clostridium *botulinum*
CA	Controlled Atmospheric Storage
CD	Crohn's disease
cDNA	Complementary DNA
CO_2	carbon dioxide
CRP	C-reactive proteins
DM	Diabetes mellitus
DNA	Deoxy nucleic acid
E.coli	Escherichia coli
EFSA	European Food Safety Authority
ELISA	Enzyme linked immunosorbent assay
GIT	gastrointestinal tract
GOS	galactooligosaccharides
H_2S	hydrogen sulfide
HAA	heterocyclic aromatic amines
HAART	Highly active antiretroviral therapy
HBV	hepatitis B virus

HDL	high-density lipoprotein
HIV	human immunodeficiency virus
HMOs	human milk oligosaccharides
IBD	inflammatory bowel disease
IBS	irritable bowel syndrome
IgE	immunoglobulin E
IL-1	interleukin-1
L. rhamnosus	Lactobacillus rhamnosus
LAB	lactic acid bacteria
LDL	low-density lipoprotein
LPS	lipopolysaccharides
MALT	Gastric mucosa-associated lymphoid tissue
MetS	metabolic syndrome
mRNA	Messenger RNA
N_2	Nitrogen gas
NAT	N-Acetyltransferase
nNRTIs	non-nucleoside reverse transcriptase inhibitors
NRTIs	nucleoside reverse transcriptase inhibitors
O_2	oxygen
PCR	polymerase chain reaction
PEF	Pulsed electric field
PIs	protease inhibitors
RA	rheumatoid arthritis
RAST	radioallergosorbent
RDA	recommended daily amount
RNA	Ribonucleic acid

RNI	reference nutrient intake
S. aureus	*staph aureus*
SCFA	short chain fatty acids
SCP	Single-cell protein
SLE	systemic lupus erythematosus
Spp	*species*
T2D	type 2 diabetes
TNF alpha	tumor necrosis factor alpha
UC	ulcerative colitis
WHO	world health organization
Y. enterocolitica	Yersinia enterocolitica

Chapter 1: Microbiota in health and disease

Objectives

Define key microbiota -related terms and concepts

Explain how the microbiome is important for maintaining human health.

Discuss role of microbiota in Disease.

Overview of Microbiota and Related Concepts

The microbiota is a collective term for the micro-organisms that live in or on the human body. Specific clusters of microbiota are found on the skin or in the gastrointestinal tract, mouth, vagina and eyes.

Human microbiome refers specifically to the collective genomes of resident microorganisms.

The Human Microbiome Project took on the project of sequencing the genome of the human microbiota, focusing particularly on the microbiota that normally inhabit the skin, mouth, nose, digestive tract, and vagina.

- The human microbiota is made up of trillions of cells - including bacteria, viruses and fungi - and they outnumber our own cells tenfold.
- The biggest populations of microbe reside in our gut - the gut microbiota. Other habitats include the skin.
- The microbial cells - and their genetic material, the microbiome - live with us in an innate relationship that is vital to normal health, although some species are also opportunistic pathogens that can invade us and cause disease.
- The microorganisms living inside the gastrointestinal tract - also known as the gut flora - amount to as much as 4 pounds of biomass, with every individual having a unique mix of species.
- The microbiota is important in nutrition, immunity and effects on the brain and behavior.
- The microbiota is implicated in numerous diseases when the normal individual balance of microbes is disturbed. Such as obesity, inflammatory bowel disease (IBD), diabetes mellitus, metabolic syndrome, atherosclerosis,

alcoholic liver disease (ALD), nonalcoholic fatty liver disease (NAFLD), cirrhosis, and hepatocellular carcinoma.

Types of microbiota:

Bacteria

Populations of microbes (such as bacteria and yeasts) inhabit the skin and mucosal surfaces in various parts of the body. Their role forms part of normal, healthy human physiology, however if microbe numbers grow beyond their typical ranges (often due to a compromised immune system) or if microbes populate (such as through poor hygiene or injury) areas of the body normally not colonized or sterile (such as the blood, or the lower respiratory tract, or the abdominal cavity), disease can result (causing, respectively, bacteremia/sepsis, pneumonia, and peritonitis).

The Human Microbiome Project found that individuals host thousands of bacterial types, different body sites having their own distinctive communities. Skin and vaginal sites showed smaller diversity than the mouth and gut, these showing the greatest richness. The bacterial makeup for a given site on a body varies from person to person, not only in type, but also in abundance. Bacteria of the same species found throughout the mouth are of multiple subtypes, preferring to inhabit distinctly different locations in the mouth.

It is estimated that 500 to 1,000 species of bacteria live in the human gut but belong to just a few phyla: Firmicutes and Bacteroidetes dominate but there are also Proteobacteria, Actinobacteria, Fusobacteria and Cyanobacteria.

Fungi

Fungi, in particular yeasts, are present in the human gut. *Candida* species due to their ability to become pathogenic in immunocompromised and even in healthy hosts. Yeasts are also present on the skin, such as *Malassezia* species, where they consume oils secreted from the sebaceous glands.

Viruses

Viruses, especially bacterial viruses (bacteriophages), colonize various body sites. These colonized sites include the skin, gut, lungs, and oral cavity.

Role of microbiota in Nutrition:

In addition to helping themselves to obtain energy from the food we eat, gut microbes are essential to the availability of nutrients for ourselves. Gut bacteria help us break down complex molecules in meats and vegetables, for example. Without the aid of gut bacteria, plant cellulose is indigestible.

Gut microbes may also have an influence on our cravings and feelings of being full after eating via their metabolic activities.

The diversity of our microbiota is related to the diversity of our diet, and adolescents trying out a wide variety of foods display a more varied gut microbiota than adults who follow a distinct dietary pattern.

Role of microbiota in Immunity

Without contact with the microorganisms that colonize us from birth, our adaptive immunity would not exist. Adaptive immunity is the part of our immune system that learns how to respond to microbes after first encountering them, enabling a more rapid defense against disease-causing organisms.

The microbiota also has relevance to autoimmune conditions and allergies, which can be more likely to develop when early microbial exposures are disturbed.

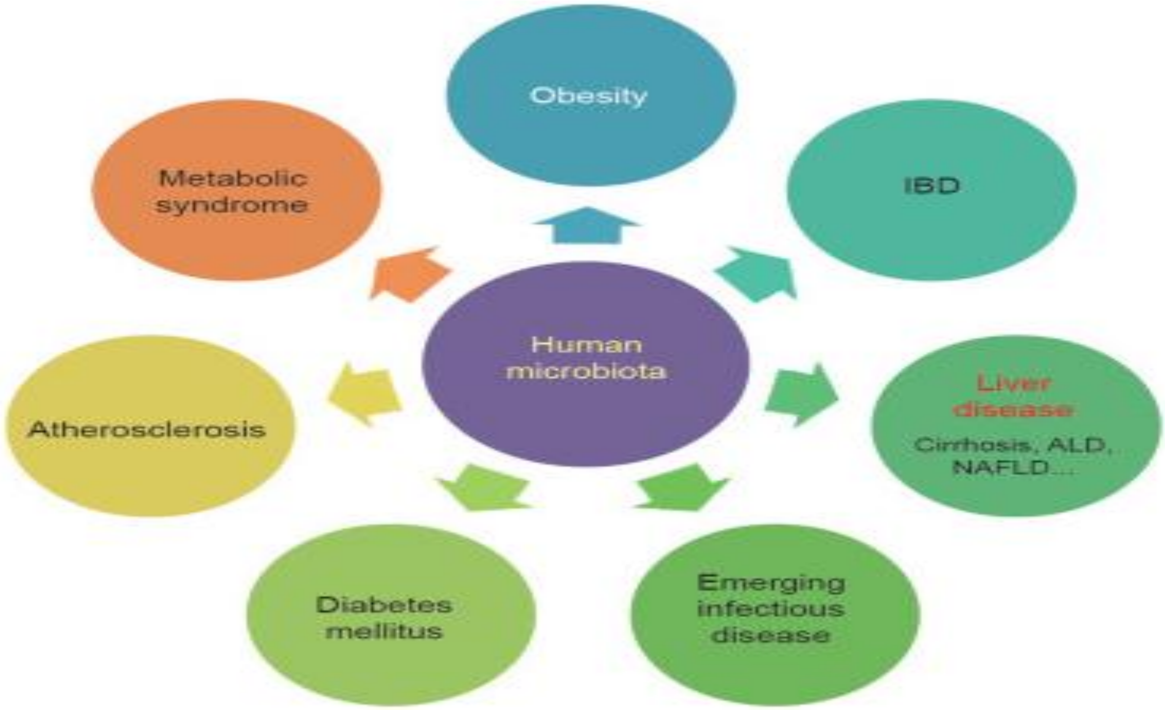
Role of microbiota in Behavior

Because of its involvement in digestion, the microbiota can also affect the brain. Some have even called the gut microbiota a "second brain."

Small molecules released by the activity of gut bacteria trigger the response of nerves in the gastrointestinal tract.

Links have also been observed between the gut microbiome and brain disorders such as depression and autistic spectrum disorder (ASD).

Role of microbiota in Disease:



The human microbiota and infectious diseases

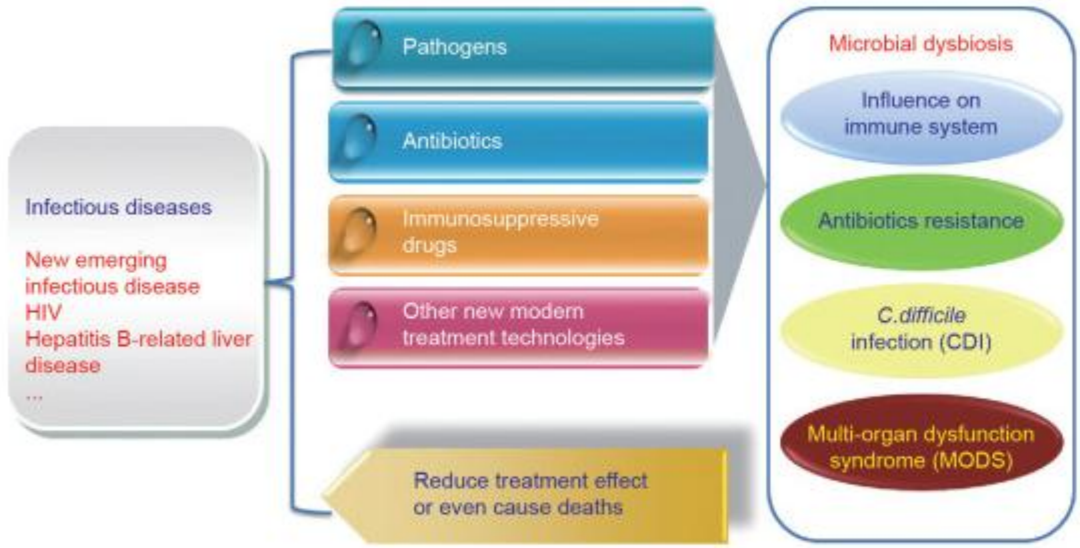
Infection is one of the most common diseases caused by dysbiosis of the microbiota. Importantly, infectious disease and its treatment have a profound impact on the human microbiota, which in turn determines the outcome of the infectious disease in the human host Disturbance of the microbiota is also associated with the progression of human immunodeficiency virus (HIV) and hepatitis B virus (HBV).

Gastrointestinal bacterial populations have provided insights into gut conditions such as the inflammatory bowel diseases (IBD) of Crohn's disease and ulcerative colitis. Low diversity in the gut microbiota has been linked to IBD as well as obesity and type 2 diabetes.

The nature of the gut microbiota has been linked to metabolic syndrome, and dietary modification has shown an effect on this cluster of risk factors via prebiotics, probiotics and other supplements.

Gut microbes and their genetics affect our energy balance, and brain development and function. As a result, research into the effects of gut microbes on the developing brain and diet-related disorders is ongoing.

Antibiotic disturbance of the microbiota can lead to disease, including the emergence of infections that display antibiotic resistance.



The innate microbiota also plays an important role in resisting intestinal overgrowth of externally introduced populations that would otherwise cause disease - the "good" bacteria compete with the "bad," with some even releasing anti-inflammatory compounds.

The human microbiota and liver diseases

The intestinal microbiota produces ethanol, ammonia, and acetaldehyde; these products may influence liver function through endotoxin release or liver metabolism .

Alterations in the intestinal microbiota play an important role in inducing and promoting liver damage progression as well as in direct injury resulting from different causal agents (e.g., viral, toxic, and metabolic agents) . through mechanisms such as the activation of liver cells by bacterial endotoxins. The gut microbiota participates in the pathogenesis of liver cirrhosis complications, such as infections, spontaneous bacterial peritonitis, hepatic encephalopathy, and renal failure. Patients with liver cirrhosis have an altered bacterial composition in their gut.

Role of microbiota in Cancer

Although cancer is generally a disease of host genetics and environmental factors, microorganisms are implicated in some 20% of human cancers. Particularly for potential factors in colon cancer, bacterial density is one million times higher than in the small intestine, and approximately 12-fold more cancers occur in the colon compared to the small intestine, possibly establishing a pathogenic role for microbiota in colon and rectal cancers. Microbial density may be used as a prognostic tool in assessment of colorectal cancers.

The microbiota may affect carcinogenesis in three broad ways:

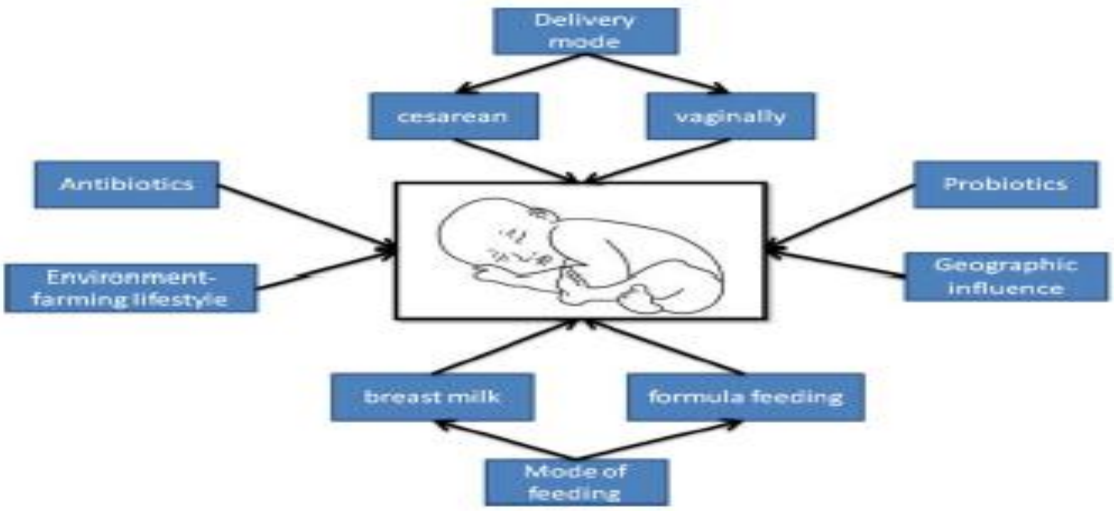
(i) altering the balance of tumor cell proliferation and death.
(ii) regulating immune system function.
(iii) influencing metabolism of host-produced factors, foods and pharmaceuticals.

Tumors arising at boundary surfaces, such as the skin, oropharynx and respiratory, digestive and urogenital tracts, harbor a microbiota. Substantial microbe presence at a tumor site does not establish association or causal links. Instead, microbes may find tumor oxygen tension or nutrient profile supportive. Decreased populations of specific microbes or induced oxidative stress may also increase risks. Microbes may secrete proteins or other factors directly drive cell proliferation in the host, or may up- or down-regulate the host immune system including driving acute or chronic inflammation in ways that contribute to carcinogenesis. Compromised host or microbiota resiliency also reduce resistance to malignancy, possibly inducing inflammation and cancer. *Helicobacter pylori* appears to increase the risk of gastric cancer, due to its driving a chronic inflammatory response in the stomach.

Practical: Gut microbiota modifications in early life

Objectives

Recognize the Early life gut microbiota.
Discuss the difference between breast milk and Formula.



Environmental factors that influence the gut microbiota.

Early life gut microbiota

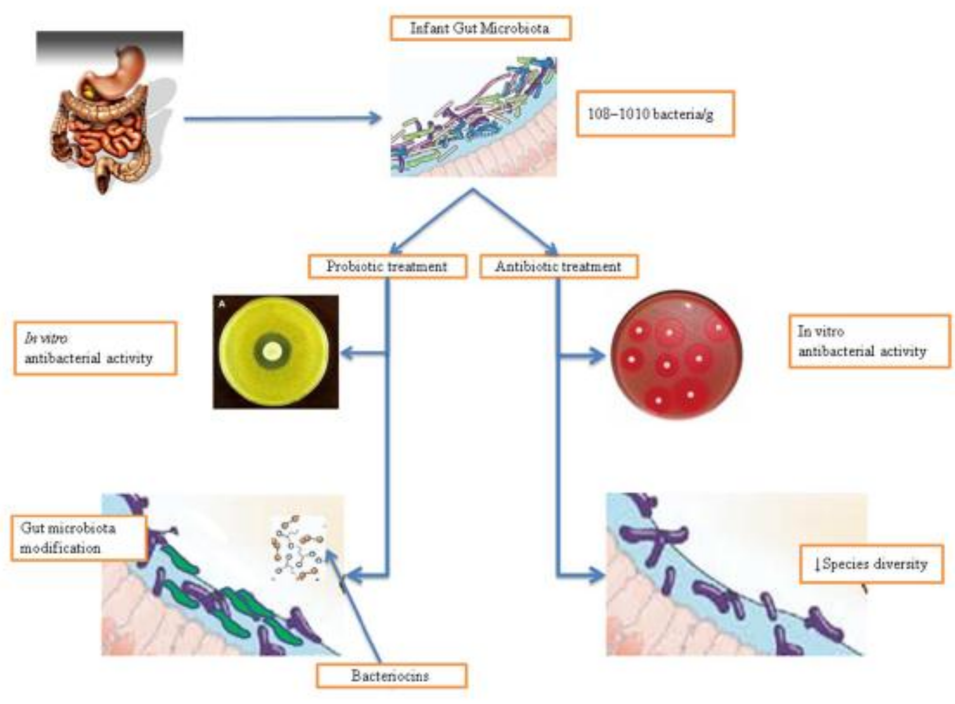
During the first days and months of life, the microbiota of the infant gut and the temporal pattern in which it evolves varies remarkably from individual to individual. At birth, infants used to be considered to be essentially free of bacteria but this was recently challenged in healthy term pregnancies, when bacteria were found in human placental membranes, amniotic fluid and umbilical cords as well as in the meconium of healthy term newborns. Immediately after birth, bacteria from the mother and/or the environment colonizes the sterile gut of a newborn infant and, within a few days, fecal density reaches 10^8–10^{10} bacteria/g. For preterm infants, composition of the gut microbiota resembles bacterial communities colonizing hospital surfaces and feeding and intubation tubing, and are enriched in Staphylococcus epidermis, Klebsiella pneumoniae, and Escherichia coli.

The early gut microbiota is often dominated by Escherichia, Clostridium, Bacteroides and Bifido bacterium.The early gut microbiome during the first days of life in infants who are born vaginally shares features with the vaginal microbial community, while in infants born by Cesarean section, the early gut microbiome resembles that of the maternal skin. Moreover, it has been proposed that the composition of the very first human microbiota could have long lasting effects on the intestine in breastfed infants.

After birth, the most important determinant of infant gut colonization is breast feeding. Human milk contains up to 10^9 live bacteria per liter and is a source of staphylococci, streptococci, lactic acid bacteria and, mostly, bifidobacterial. Among breastfed infants, bifidobacterial and particularly B. longum, B. infantis and B. breve can reach up to 60–90% of the total fecal microbiota. Moreover, the gut microbiota of exclusively breastfed infants exhibits significant differences in gut microbiota as compared to that of formula fed infants.

Formula fed microbiota is more complex and is characterized by the predominance of facultative anaerobes such as Bacteroides and Clostridium followed by Staphylococcus, Streptococcus and Enterobacteriaceae, and delayed colonization by bifido bacteria.

After the first six months of life, when solid food is introduced to the infant, the gut microbiota becomes more diverse and the abundance of Bacteroides, Clostridium and anaerobic bacteria increases rapidly while the proportion of bifido bacteria becomes more stable.



Gut microbiota modifications after the administration of probiotics and antibiotics.

Probiotics in early life

Probiotics have increasingly been administrated to children. Bifidobacterium lactis (B. lactis) probiotics beyond early infancy are associated with a reduced risk of nonspecific gastrointestinal infections in children.

Probiotics and weight gain

The perinatal use of Lactobacillus rhamnosus (L. rhamnosus) suggest that early modulation of the gut microbiota modifies children's growth patterns by inhibiting excessive weight gain during the first few years of life.

Healthy infants who received Lactobacillus rhamnosus enriched formula during the first six months of life presented significantly increased body length and weight. The administration of Bifidobacterium breve probiotics to very low birth weight infants was associated with improved weight gain as well as a more rapid growth of Lactobacillus as compared to infants who did not receive probiotic Supplements.

Moreover, probiotic treatment of very low birth weight infants with Lactobacillus acidophilus and Bifidobacterium infantis has been shown to reduce morbidity as well as to increase daily weight gain and decrease the length of hospital stay.

Probiotics have also been used to treat malnutrition in children: ready-to-use therapeutic food has reduced the prevalence of malnutrition and led to weight gain in children.

So early modulation of the gut microbiota through the administration of probiotics is likely to result in increased weight gain.

Antibiotics in early life

Exposure to antibiotics remains very high perinatally and in the first periods of life. A number of neonates, particularly premature infants, receive antibiotics to prevent or treat bacterial infections. Moreover, many mothers receive antibiotics for prophylaxis of vaginal group B streptococcus and Cesarean section delivery. Antibiotic treatment has been associated with disturbances of the gut microbiota in the actively developing infant gut microbiota.

Antibiotic treatment during the first two years of life decreases gut flora biodiversity.

Early life antibiotic treatment has been linked with an increased risk of obesity and related metabolic sequelae later in life.

Chapter2: The Gut Microbiota

Objectives

Recognize Interaction among gut microbiota, host and food.
Explain The role of gut microbes on the digestion of macronutrients.
Discuss impact of diet on GIT microbiota.

Overview of gut microbiota and Related Concepts

The gut microbiota is an ecological community of symbiotic, commensal and pathogenic microorganisms. This microbial ecosystem includes many bacterial species which permanently colonize the gastrointestinal tract as well as a large number of microorganisms such as Archaea, viruses, parasites and fungi that come from our environment. The number of these microorganisms can reach 10^{12}–10^{14} in the colon, making gut microbiota one of the most densely populated communities, far exceeding that of the soil, the subsoil, and the oceans.

In a healthy human adult, the gut microbiota

The dominant bacterial species in the human gastrointestinal tract are divided into three phyla:

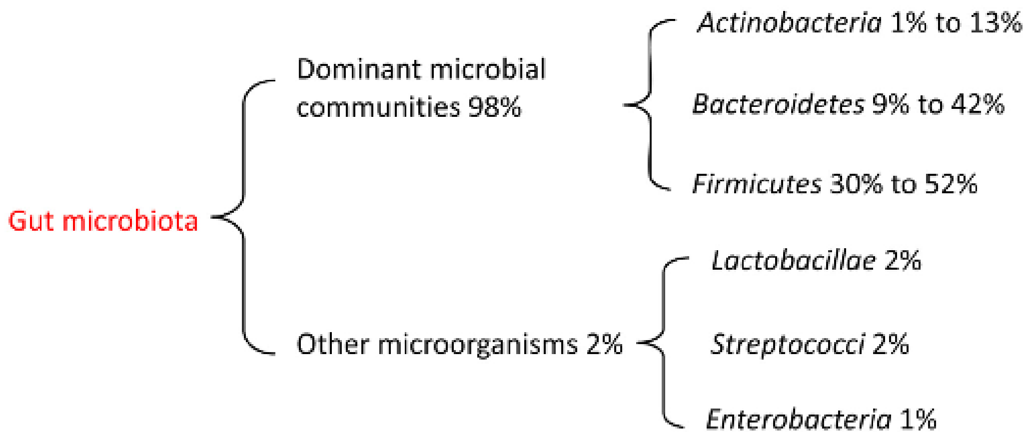
The phylum Bacteroidetes (e.g. Porphyromonas, Prevotella etc.),

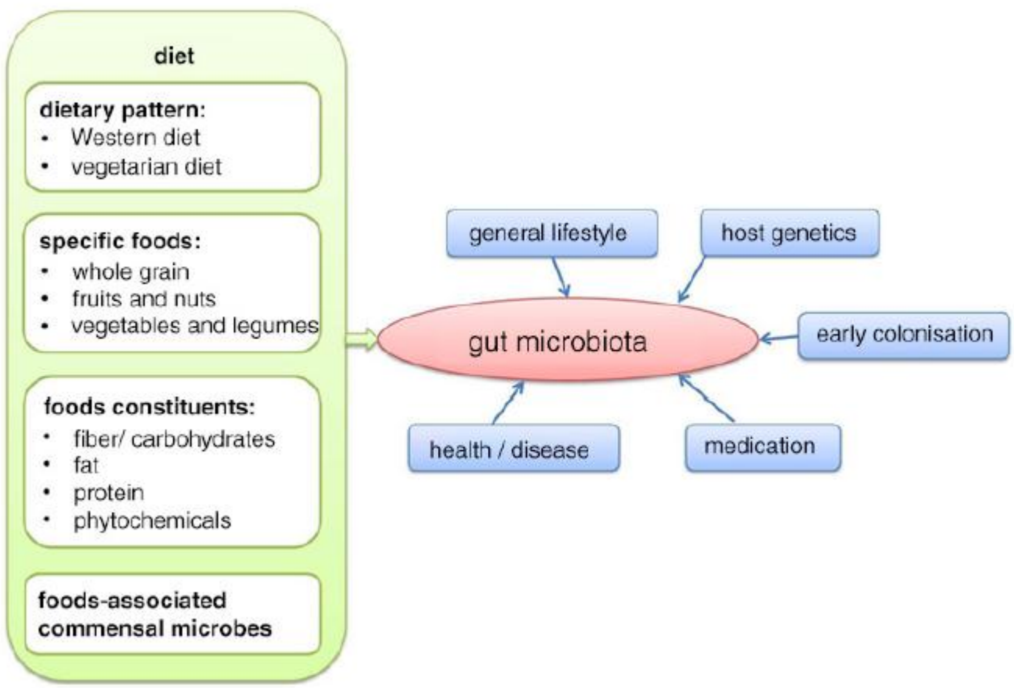
The phylum Firmicutes (e.g. Ruminococcus, Clostridium, Eubacteria etc.)

And the phylum Actinobacteria (Bifidobacterium). Other bacteria such as Lactobacilli, Streptococci and Escherichia coli (E.coli) are found in small numbers.

The Bacteroidetes and Firmicutes phyla were found to be the dominant bacterial

Populations in the gastrointestinal tract (GIT).



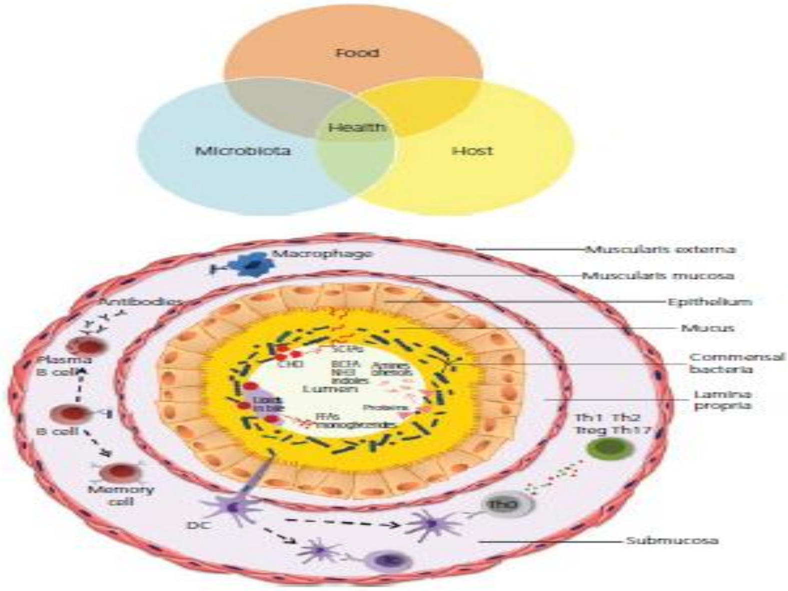


Factors, which influence the composition of the human gut microbiota, with special focus on diet.

Interaction among gut microbiota, host and food

The intestinal bacteria reside at the interface of the external environment and the host, allowing for a tightly regulated crosstalk between the three. The bacteria reside within the lumen of the intestinal tract and interact directly with dietary antigens where they aid in digestion of partially digested food, vitamin biosynthesis, and absorption. The bacteria also act as a first line of defense against pathogens in the gut through competition for food and space.

interactions among the intestinal microbiota, host, and the environment regulate intestinal health and by extension the host. Disturbances in any of these factors can have deleterious effects on the host.



The role of gut microbes on the digestion of macronutrients

To utilize food energy, the host-microbiome superorganism has evolved to work as a unit customized to the benefit of both. Up to 10% of the dietary energy in humans can be due to the activities of their intestinal microbiota. This may be beneficial or detrimental depending on the needs of the host. For example, in the context of obesity, having microbes that extract more energy is not favorable but in the context of cachexia this would be of benefit.

Carbohydrates

Mammalian genomes do not encode for most enzymes required for the degradation of insoluble structural polysaccharides. Undigested food particles arriving at the large intestine are typically comprised of insoluble materials derived from plant cell walls and resistant starch. Specialized bacteria species such as *Roseburia bromii*, belonging to the Firmicutes, act as primary degraders of insoluble polysaccharides, in turn producing substrates for other amylolytic bacteria. The primary result of carbohydrate fermentation under anaerobic conditions in the gut is the production of short-chain fatty acids (SCFAs) such as acetate, lactate, propionate, and butyrate, of which the last plays a particularly important role in gut homeostasis and health. While lactate and acetate become available to the host systemically, butyrate directly becomes the primary food source of the colonocytes. Butyrate is known to possess anti-cancer and anti-inflammatory properties43 and is involved in gut motility, energy expenditure, and appetite control. Butyrate can also be produced through conversion of other available acids such as acetate and lactate by members of the Firmicutes phyla, including *Eubacterium hallii* and *Anaerostipes* spp.

Proteins

Partially digested proteins, mucus secretions, and amino acids from shed IEC are fermented in the large intestine and serve as a source of carbon and nitrogen for intestinal microorganisms. The amount of protein entering the large intestine depends on the total intake as well as the source of the protein. The digestibility of animal-derived protein is higher (94–99%) than that derived from plant sources (70–90%)50. Protein catabolism by intestinal microorganisms yields a variety of end products, many which are toxic to the host. Degradation of undigested or endogenous protein, referred to as putrefaction, is particularly prominent in the etiology of ulcerative colitis (UC). Bacterial proteases and peptidases can initiate the degradation process by means of hydrolysis to convert proteins into smaller peptides and amino acids. This process is optimal in the distal colon where pH is higher. Fermentation of amino acids by reductive deamination can, albeit to a much lesser extent, produce short chain fatty acids (SCFA) similar to those found during carbohydrate fermentation. They also produce ammonia, amines, thiols, phenols, and indoles, which are exclusive to amino acid fermentation. The accumulation of these by-products is pathogenic to the host. For example, phenols and indoles are considered carcinogens, ammonia a mutagen, and thiols a cellular toxin. Another product of protein fermentation is branched-chain fatty acids (BCFA). The physiological significance of BCFA is not well understood; however, they appear to be important in developmental stages during gestation and immediately following birth. Further, sulfate-reducing bacteria such as those in the genus *Desulfovibrio* instigate fermentation of dietary and mucinous sulfate and sulfur amino acids, leading to increased production of hydrogen sulfide (H_2S), which has been associated with pathogenesis of various intestinal diseases including ulcerative colitis. As H_2S is involved in many normal processes within the colon, it is likely that its association with etiology of intestinal diseases is relevant only in conditions where H_2S levels reach higher than subtoxic levels.

Lipids

Different members of the gut microbiota are thought to regulate fat absorption via distinct mechanisms. Several roles of microbes in the harvest and storage of dietary lipids have been postulated; however, the exact mechanisms in play remain to be elucidated.

Once dietary lipids are absorbed into the enterocytes, they are repackaged into lipoprotein particles known as chylomicrons. These chylomicrons transport lipids in the form of triglycerides to adipocytes, where lipoprotein lipases breakdown triglycerides into free fatty acids that can then be taken into the tissue for storage. the presence of gut microbes reduced serum levels of chylomicrons.

This outcome could have been due to decreased lipid absorption in the gut or an increased lipid clearance in the periphery. So the presence of microbiota did not affect the absorbance rate of lipid, but rather affected the clearance of chylomicrons.

Impact of diet on GIT microbiota

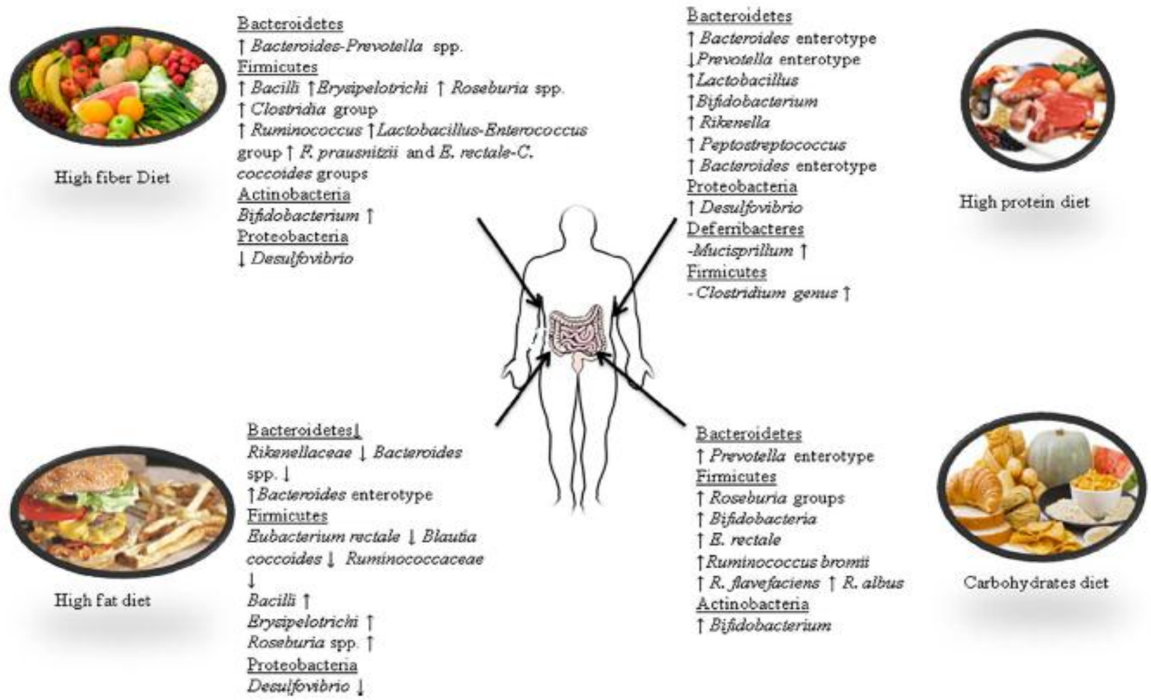


Fig. 1. Influence of the main dietary components in gut microbiota composition.

It has been widely reported that the consumption of **modern western diets** containing less fiber and vegetables tended to result in the loss of some important microbial species in the western (urban) communities compared to rural communities. comparing an individual whose diet is high in fat and low in fiber with an individual on the opposite diet (*e.g.* high fiber and low fat), the latter tends to have a smaller amount of pathogenic bacteria and a large amount of beneficial microbes, such as *Pre-votella* and *Xylanibacte*. The balance of GIT microbial composition can be achieved as a result of symbiosis which regulates the immune system and protects the host from various diseases.

The Mediterranean diet, which is based on a balanced intake of fruits, grains, monounsaturated fat, vegetables and polyunsaturated fats, is considered the standard for a healthy life style. It has been found that such diets have anti-inflammatory capabilities and can be used to reduce inflammation in diseases. Individuals fed on the Mediterranean diet have lower numbers of *Bacillaceae*, *Proteobacteria* and acute phase C-reactive proteins (CRP), but higher *Clostridium* and *Bacteroidetes* populations.

Vegetarian diets are also recognized as healthy and benefic diets because they can protect the host from various chronic, metabolic and inflammatory disorders. Recent investigations showed that vegetarian diets could increase the number of *Faecalibacterium praus-nitzii*, *Clostridium clostridioforme* and *Bacteroides Prevotella*,but decrease the ratio of *Clostridium cluster* species. Small amounts of dietary fat can be digested and absorbed but some fat components cannot be metabolized and pass to the colon where they affect the microbial composition and are then excreted in feces. Consequently, the consumption of high-fat foods tends to induce substantial changes in the composition of GI tract microbiota. High fat content and increased calorie consumption have the capability to induce changes in the microbial composition of the GI tract. It has been also found that the children feeding on vegetarian diets rich in plant-based polysaccharides, fibers and starches had a significant increase in the number of *Firmicutes*, *Xylanibacter*,*Bacteroidetes* and *Prevotella* compared with those consuming a carbohydrate rich European diet. Therefore, it was suggested that children should be provided with a plant-based polysaccharide rich diet which confers protection against inflammatory disease Currently, it is clear that the composition of the microbiota differs among individuals living in different geographic regions and also depends on the long-term diet pattern.

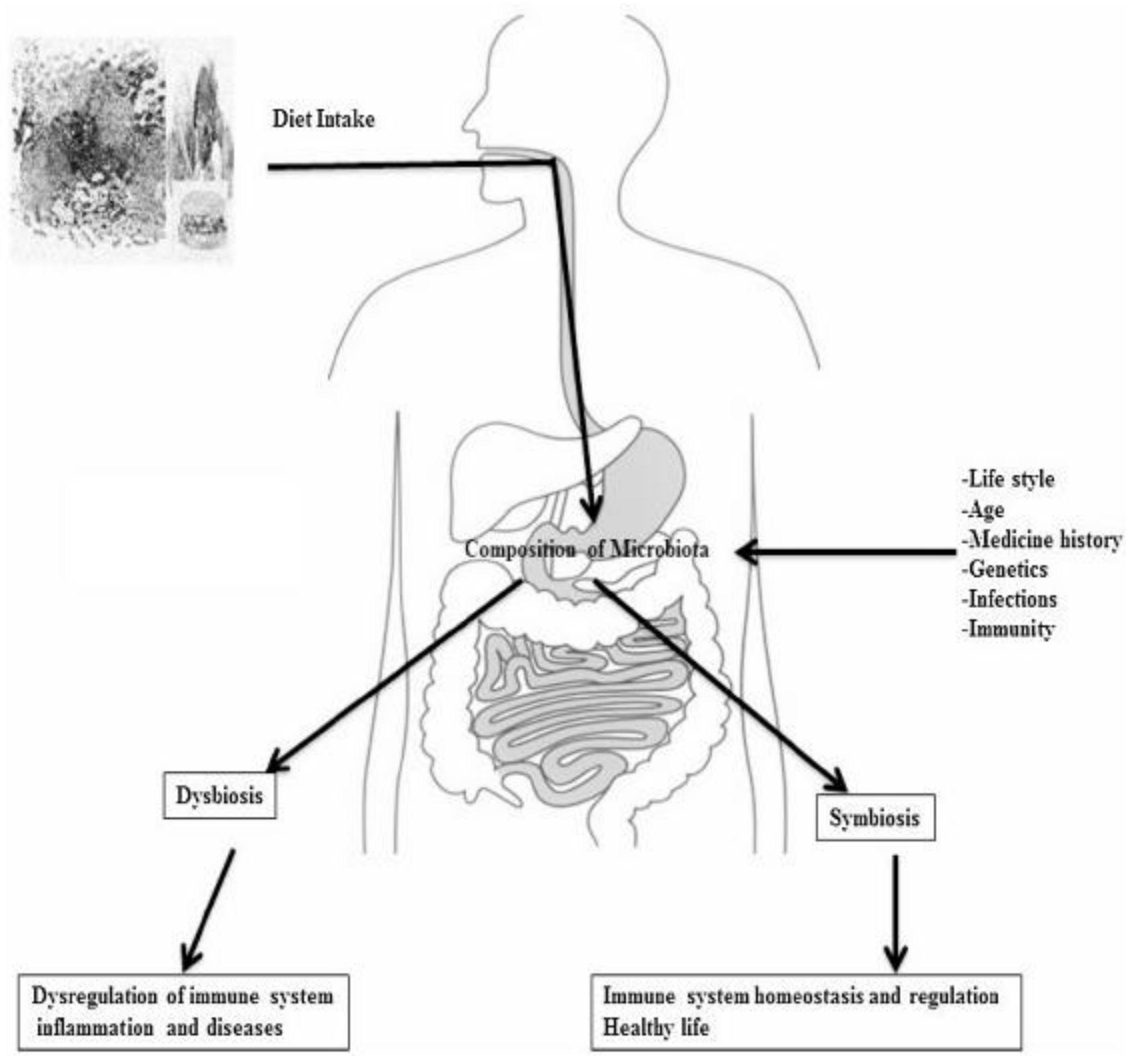
Practical: The gut microbiota role in obesity and diabetes

Objectives

Recognize obesogenic microbiota in humans.

Discuss how Obesity-proneness: mediated by the gut microbiota.

Recognize the role of microbiota in diabetes.



Microbiota and homeostasis of the immune system.

the metabolic syndrome (MetS) includes increased waist circumference, hyperglycemia, elevated blood pressure and hyperlipidemia. With time, these conditions present a major risk of developing obesity, type 2 diabetes and atherosclerosis. To date, treatment mainly includes

symptom management, and effective prevention strategies are largely lacking.With the exciting discovery of an “obesogenic” microbiota in recent years, with attempts to use microbial manipulations to modulate our gut microbiome as a new preventive and therapeutic approach for different aspects of the MetS.

The "obesogenic" microbiota in humans

In humans, clear shifts in the gut microbiome can be observed when comparing obese and lean individuals, with increased *Firmicutes* to *Bacteroides* ratio in obese individuals. Nutrient content in the diet directly affects the gut microbiome, where high-energy diets increase the ratio of *Firmicutes* to *Bacteroides* in humans. Thus, a diet high in energy will contribute to development of obesity not only due to the energy content in the diet, but also through maintaining an "obesogenic" gut microbiota.

It is clear that a high-fat diet induces an "obese" microbiota independent of body weight state and the altered gut microbiota in obese individuals is not merely a consequence of the obese state.

A leaky gut contributing to inflammation and adiposity

The monolayer of epithelial cells lining the gut mucosa has the delicate dual function of being an efficient absorptive layer to nutrients, whilst still maintaining a tight barrier to invading pathogens. This is accomplished through several specific features of the intestinal epithelium: first, the intestine hosts 70–90% of the body's immune cells, which together with the extensive enteric nervous system closely monitor events in the gut. Second, the junctions between epithelial cells consist of an intricate collection of proteins comprising the so-called tight junctions, which can be regulated by the cells themselves but are also affected by bacteria. Similarly, in obesity, these junctions appear leaky, as an increased influx of bacterial cell wall lipopolysaccharides (LPS) is observed as compared to lean individuals. LPS is a component of Gram negative bacteria cell walls and is pro-inflammatory substance.The low-grade systemic inflammation observed in obese individuals can be ascribed partly to increased LPS levels in the blood, but also to an increased secretion of pro-inflammatory cytokines from adipose tissue. Interestingly, a high-fat diet directly increases LPS blood levels, as LPS can form and bind to micelles during fat absorption and are therefore shuttled into the body from the gut.

Obesity-proneness: mediated by the gut microbiota

In humans resistance to diet-induced obesity is occasionally observed, but the reasons remain unclear why the same amount of energy ingested does not result in the same weight increase in all individuals. In obesity-prone and obesity-resistant mice fed a high-fat diet, fecal carbohydrate calories differed, whereas fecal fat or protein calories did not differ. This surprisingly indicates that lean person display decreased carbohydrate absorption from the diet as compared to obese person, despite similar caloric intake. Carbohydrate absorption is greatly affected by gut microbial activities, and there is differences in microbial composition between obesity-prone and obesity-resistant persons.

Bacterial metabolites provide a link between bacteria and host metabolism

The gut bacteria degrade dietary component sreaching the colon, giving rise to metabolites such as short-chain fatty acids (SCFA), which are formed during bacterial digestion of indigestible carbohydrates (e.g. dietary fiber). These bacterial metabolites may play an important role for host physiology, as they can be utilized as energy by colon cells, as well as be absorbed into the blood stream. Thus, a diet low in dietary fiber may affect colonic health.

The "diabetic" microbiota

Diabetes mellitus (DM) presents in two major forms: type I and type II, characterized by vastly different molecular events leading up to malfunction of glucose homeostasis. In type I diabetes, autoimmune reactions to insulin-producing beta-cells in the pancreas results in cell death and a gradual loss of insulin-production capacity, often starting early in life. The disease requires careful monitoring of blood glucose levels and life-long administration of insulin. Type II diabetes, on the other hand, is usually described as life-style-related, as obesity, little exercise and an unhealthy diet are key risk factors. Type II diabetics can often reduce their need for insulin by adopting a more healthy life style. Another important difference between the two forms of diabetes is the phenomenon of "insulin resistance" in type II diabetics. These patients have no lack of insulin; instead, they become insensitive to insulin and require higher doses of insulin to be able to transport glucose from the blood stream into the cells.

Type I diabetes and the gut microbiota

It is currently unknown what triggers the autoimmune reactions in type I diabetes, Bacteria in the phylum *Bacteroidetes* (including *Prevotella* and unclassified *Bacteriodales*) may protect against development of type I diabetes, while increased levels of members of the *Firmicutes* phylum, i.e. *Ruminococcus*, *Oscillospira* and *Lachnospiraceae*, promoted the disease. Type I diabetic children were also characterized by increased gut microbial diversity. This is interesting, as obese and type II diabetic adults generally have lower gut microbial diversity, which may point towards a complex age-related dynamic of the gut microbiota development.

Type II diabetes

Diabetes Type 2 alters the composition of gut microbiota and microbiota function such as the secondary metabolite bile acid and butyrate products. These functions are crucial for insulin sensitivity. The status of gut microbiota can be used to success-fully distinguish between type 2 diabetes patients and healthy individuals. Type 2 diabetes was linked to higher amounts of Lactobacilli and lower amounts of Roseburia when comparing the two populations.

Chapter 3: The microbiota in inflammatory bowel disease

Objectives

Explain Dysbiosis and IBD pathogenesis.
Recognize Nutritional and dietetic therapies

Overview of inflammatory bowel disease and Related Concepts

Inflammatory bowel disease (IBD) comprises two distinct conditions, ulcerative colitis (UC) and Crohn's disease (CD) that are characterized by chronic relapsing inflammation of the gut in genetically susceptible individuals exposed to defined environmental risk factors.

Dysbiosis and IBD pathogenesis

The relationship between the gut commensal microbiota and IBD is complex and can be explained by four broad mechanisms:
(i) conventional microbiota dysbiosis,
(ii) intestinal inflammation induction by pathogens and/or functionally altered commensal bacteria.
(iii) genetic defects in the host resulting in the loss of the commensal microbiota.
(iv) defective immunoregulation in the host.

Causes

IBD is a complex disease which arises as a result of the interaction of environmental and genetic factors leading to immunological responses and inflammation in the intestine.

Diet

Dietary patterns are associated with a risk for ulcerative colitis. In particular, subjects who were in the highest tertile of the healthy dietary pattern had a 79% lower risk of ulcerative colitis. Gluten sensitivity is common in IBD and associated with having flareups. A diet high in protein, particular animal protein, may be associated with increased risk of inflammatory bowel disease and relapses.

Microbiota

As a result of microbial symbiosis and immunity, alterations in the gut microbiome may contribute to inflammatory gut diseases. IBD-affected individuals have been found to have 30–50 percent reduced biodiversity of commensal bacteria, such as decreases in Firmicutes (namely Lachnospiraceae) and Bacteroidetes. Further evidence of the role of gut flora in the cause of inflammatory bowel disease is that IBD-affected individuals are more likely to have been prescribed antibiotics in the 2–5 year period before their diagnosis than unaffected individuals. The enteral bacteria can be altered by environmental factors, such as concentrated milk fats (a common ingredient of processed foods and confectionery) or oral medications such as antibiotics and oral iron preparations.

Breach of intestinal barrier

Loss of integrity of the intestinal epithelium plays a key pathogenic role in IBD. Changes in the composition of the intestinal microbiota are an important environmental factor in the development of IBD. Detrimental changes in the intestinal microbiota induce an inappropriate immune response that results in damage to the intestinal epithelium. Breaches in this critical barrier (the intestinal epithelium) allow further infiltration of microbiota that, in turn, elicit further immune responses. IBD is a multifactorial disease that is nonetheless driven in part by an exaggerated immune response to gut microbiota that causes defects in epithelial barrier function.

Nutritional and dietetic therapies

Nutritional deficiencies play a prominent role in IBD. Malabsorption, diarrhea, and GI blood loss are common features of IBD. Deficiencies of B vitamins, fat-soluble vitamins, essential fatty acids, and key minerals such as magnesium, zinc, and selenium are extremely common and benefit from replacement therapy. Dietary interventions, including certain exclusion diets, low fiber diets has some benefits.

Microbiome

There is evidence of an infectious contribution to inflammatory bowel disease in some patients and this subgroup of patients may benefit from antibiotic therapy.

Fecal microbiota transplant is a relatively new treatment option for IBD. Patients benefits similar to those in Clostridium difficile infection.

*Around one-third of individuals with IBD experience persistent gastrointestinal symptoms similar to irritable bowel syndrome (IBS) in the absence of objective evidence of disease activity. The cause of these IBS-like symptoms is unclear, but it has been suggested that changes in the gut-brain axis, epithelial barrier dysfunction, and the gut flora may be partially responsible. Patients of IBD do have an increased risk of colorectal cancer.

Practical: The main dietary goals in the management of IBD

Objectives

Discuss main reasons for malnutrition in patients with inflammatory bowel disease.
Recognize the main nutritional deficiencies of inflammatory bowel disease.
Recognize main dietary goals in the management of Crohn’s disease.

What are the main reasons for malnutrition in patients with inflammatory bowel disease?

Several mechanisms contribute to the malnutrition observed in inflammatory bowel disease (IBD) patients. A decrease in the oral intake of nutrients is a common symptom and is often due to abdominal pain, diarrhoea, anorexia, nausea and vomiting and in cases of strictures or abscesses. Moreover, mucosal inflammation and its associated diarrhoea or bleeding lead to a loss of protein, blood, minerals, electrolytes and trace elements. Multiple resections, bacterial overgrowth and fistulas between the small and large intestine may have an adverse nutritional effect (e.g. vitamins and minerals) owing to a decreased absorptive area and subsequent malabsorption. Furthermore, increased energy requirements because of inflammation or fever can lead to weight loss and altered intermediate metabolism, owing to increased tumor necrosis factor alpha (TNF alpha), interleukin-1(IL-1) and 6, which in turn lead to decreased albumin and other protein synthesis. Pharmacological therapies may also lead to malnutrition. For example, sulfasalazine reduces folic acid absorption, whereas corticosteroids decrease calcium absorption and negatively affect protein metabolism. Lastly, restrictive diets that are recommended to patients by family, friends and physicians or prolonged periods of fasting because of exacerbations or diagnostic procedures also contribute to the risk of malnutrition. The consequences of malnutrition are numerous, and include reductions in bone mineral density, as well as growth retardation and delayed sexual maturity in children. Osteoporosis may also be implicated as a result of pro-inflammatory cytokine profiles.

What are the main nutritional deficiencies of inflammatory bowel disease?

IBD is associated with a number of nutritional deficiencies, including anaemia, hypoalbuminaemia, hypomagnesaemia, hypocalcaemia and hypophosphataemia, as well as deficiencies in folic acid, niacin, fat-soluble vitamins and B12 in cases of terminal ileum resection and deficiencies of iron, zinc and copper. Plasma antioxidant concentrations are also reduced in many IBD patients, particularly those with active disease. Patients with diarrhoea and vomiting may also experience electrolyte imbalances.

What kind of diet should be administered to a patient with ulcerative colitis exacerbation?

For nutritional support, enteral feeds are generally preferred to parenteral nutrition, except from cases of toxic megacolon, extended colon haemorrhage, perforation or obstruction, which require bowel rest and parenteral administration of fluids and nutrients. In times of exacerbation, a liquid diet is first administered, followed by a low-residue diet. In patients with strictures, a low-fibre diet is required in order to prevent obstruction. When the patient enters the remission phase, they should be encouraged to consume a variety of foods from all food groups and dietitians should help patients realise their own intolerances and further compose a balanced diet.

Which are the main dietary goals in the management of Crohn's disease?

Medical nutritional therapy for patients with Crohn's disease (CD) aims to:

_ prevent or restore protein/energy malnutrition
_ assess and correct micronutrient deficiencies
_ maintain bowel rest in periods of exacerbation
_ modify the diet regime according to drug treatment and drug–nutrient interactions
_ modulate immune response by modulating cytokines expression (e.g. omega-3 polyunsaturated fatty acids), by reducing gut permeability and enhancing gut barrier (e.g. probiotics), by affecting gene expression and by modulating local immunity in the intestine (e.g. butyrate, glutamine).

Chapter4: Food microbiology and food safety

Objectives

Recognize food safety related concepts.
List the commonest causative agents of foodborne illness.
Recognize factors affecting microbial growth.
Discuss Microbiological Testing of food.

Overview of food microbiology and Related Concepts

Food microbiology is the study of the microorganisms that inhabit, create, or contaminate food. Of major importance is the study of microorganisms causing food spoilage. "Good" bacteria, however, such as probiotics, are becoming increasingly important in food science. In addition, microorganisms are essential for the production of foods such as cheese, yogurt, and other fermented foods such as bread, beer and wine.

Food safety is a major focus of food microbiology. Pathogenic bacteria, viruses and toxins produced by microorganisms are all possible contaminants of food. However, microorganisms and their products can also be used to combat these pathogenic microbes. Probiotic bacteria, including those that produce bacteriocins, can kill and inhibit pathogens. Alternatively, purified bacteriocins such as nisin can be added directly to food products. Finally, bacteriophages, viruses that only infect bacteria, can be used to kill bacterial pathogens. Thorough preparation of food, including proper cooking, eliminates most bacteria and viruses. However, toxins produced by contaminants may not be heat-labile, and some are not eliminated by cooking.

Fermentation is one-way microorganisms can change a food. Yeast, especially Saccharomyces cerevisiae, is used to leaven bread, brew beer and make wine. Certain bacteria, including lactic acid bacteria, are used to make yogurt, cheese, hot sauce, pickles, fermented sausages and dishes such as kimchi. A common effect of these fermentations is that the food product is less hospitable to other microorganisms, including pathogens and spoilage-causing microorganisms, thus

Extending the food's shelf-life. Food fermentations are ancient technologies that harness microorganisms and their enzymes to improve the human diet. Fermented foods keep better, have enhanced flavours, textures and aromas, and may also possess certain health benefits, including superior digestibility. For vegetarians, fermented foods serve as palatable, protein-rich meat substitutes. Some cheese varieties also require molds to ripen and develop their characteristic flavors.

Microorganisms associated with foods

can be categorized as "spoilage," "pathogenic," or "useful." Spoilage microorganisms are those that can grow in a food and cause undesirable changes in flavor, consistency (body and texture), color, or appearance. Also bacterial enzymes may effect slow deterioration of frozen or dried foods during long-time storage. These changes diminish the quality characteristics of foods and may render them ultimately unfit for human consumption. For example, refrigerated perishable foods such as milk, fresh meat, poultry, fish, fruits, and vegetables lose some quality characteristics during normal storage and ultimately spoil, due in part to the activity of microorganisms capable of growth at refrigeration temperatures. Usually, extensive microbial growth (millions of organisms per g or cm2) occurs before quality losses are perceptible. These changes, when perceived by the consumer, serve as an alert that extensive microbial activity has taken place. Pathogenic microorganisms can render foods harmful to humans in a variety of ways. Foods may serve as the vehicle for the introduction of infectious
microorganisms into the gastrointestinal tract, e.g., *Salmonella* and *Shigella*. Multiplication of certain microorganisms in foods prior to consumption may result in production of toxins, e.g., *Clostridium botulinum*, *Staphylococcus aureus*, and *Bacillus cereus*. Foods may also be the vehicle for microorganisms that form toxins *in vivo*, e.g., *Clostridium perfringens* and certain pathogenic *Escherichia coli*.

With some foods, conditions are chosen to favor the development of useful microorganisms such as lactic acid bacteria and yeasts, which are either naturally present or added intentionally. Such foods as cheeses, yogurt, breads, pickles, and fermented sausages offer desirable organoleptic properties and shelf-life.

Food as a Selective Environment

Microbial activities in foods can be viewed from the perspective of the food as a "selective environment," despite the diversity of microorganisms that contaminate the surfaces of the raw materials. The selectivity is imposed by the physical-chemical characteristics of the food, the additives it contains, the processing techniques, the packaging material, and the storage conditions. It is necessary to distinguish between the shelf-life of two broad categories of foods, namely those that are shelf-stable and those that are perishable. shelf-life will be treated as it relates to microbial activity only.

Microbiological shelf-stability of many foods is related to storage conditions. For example, dried and frozen foods are microbiologically shelf stable as long as they remain dry or frozen. Shelf-stable foods are not necessarily sterile; indeed, many do contain microorganisms. Some shelf-stable canned foods may undergo microbiological spoilage if they are exposed to elevated temperatures permitting the growth of surviving thermophilic spore forming bacteria, whereas these organisms are inactive at ambient temperatures and indeed tend to die during normal storage. Shelf-stable food is distinguished from perishable food in that an attribute or attributes of the shelf-stable food prevent the growth of contaminating microorganisms. For example, certain canned products are heat processed to the degree that they are sterile; the attribute assuring stability of such products is elimination of all living forms. With many shelf-stable foods, other attributes prevent microbial growth. Dried beans are shelf-stable because they contain insufficient moisture to permit microbial growth. Mayonnaise is shelf-stable because it contains sufficient quantities of acetic acid in the moisture phase of the product to prevent growth of contaminating organisms. Certain canned cured meats are shelf-stable, not because they are sterile, but because sublethal heat treatment so injures surviving spores that they are incapable of outgrowth in the presence of salt and nitrite. The distinguishing characteristic of shelf-stable foods, then, is their resistance to microbiological spoilage. Microbial growth in such products is an abnormal and unexpected event.

Perishable foods, on the other hand, have a finite shelf-life and if not consumed, will spoil at some time during storage. The exact time of spoilage depends upon a great number of variables. Though various processing procedures, additives, packaging methods, and storage conditions may be applied to increase shelf-life, microorganisms capable of growth survive and ultimately grow. When such growth proceeds to the extent that undesirable changes are perceptible to the processor, preparer, or consumer, the food is deemed of inferior quality or spoiled and is rejected. The distinguishing feature of perishable foods, in contrast to shelf-stable foods, is that microbiological spoilage is an expected event. It will ultimately occur even if the food has been prepared from wholesome raw materials and has been properly processed, packaged, and stored.

Microflora of Processed Foods

Although the microflora of raw materials is usually heterogeneous, processing of foods (except those that are sterile) often imposes a characteristic and highly specific microbiological flora. The normal flora of severely heat processed, but not sterilized, low-acid canned foods is comprised of thermophilic spore forming bacteria, the most heat-resistant microbial components of the raw materials.

The predominating flora of shelf-stable canned cured meats consists of mesophilic aerobic and anaerobic spore forming bacteria, the predominant organisms resistant to the heat process applied to these products. The normal flora of mayonnaise and salad dressing is comprised of small numbers of spore forming bacteria, yeasts, and lactic acid bacteria. Aerobic spore forming bacteria predominate in dry spices and in a number of dry vegetable products. Molds and yeasts predominate in dried fruits. The normal flora in carbonated beverages is comprised of yeasts. In each of the foregoing, the surviving and predominating microflora reflects the nature of the raw materials, processing conditions, packaging, and storage of the shelf-stable product.

However, spoilage is still possible. If the severely heat-processed canned foods were exposed to high temperatures during storage, spoilage due to the germination and outgrowth of thermophilic spore forming bacteria might occur. If shelf-stable canned cured meat were to contain excessive numbers of aerobic spore forming bacteria, growth of these organisms might result in spoilage, despite an adequate heat process and normal levels of salt and nitrite. Excessive levels of yeasts or lactic acid bacteria might result in their growth and subsequent spoilage of the mayonnaise, despite levels of acetic acid that would assure the stability of a product containing "normal" levels of the same organisms. Time/temperature abuse of an ingredient of a carbonated beverage (for example, a flavor) may lead to the development of large numbers of yeasts that could overcome the effect of carbonic acid, which would normally render the same beverage stable.

The normal flora in microbiologically shelf-stable products is, therefore, rather specific. If the stabilizing nature of the system should be overcome, this microflora may multiply and cause spoilage—an unexpected event. With perishable products, the microflora that survives processing may be heterogeneous, but that portion of it developing during storage and causing spoilage is usually quite specific. For example, a heterogeneous flora exists on raw red meats, poultry, and fish as a result of contamination from the animal and/or the processing environment. Yet, during refrigerated storage of such products, spoilage is caused predominantly by a highly specific group of microorganisms, namely *Pseudomonas* and closely related aerobic, psychrotrophic gram-negative bacteria. If the same products are vacuum-packed in oxygen-impermeable films, a different microflora becomes predominant, namely, lactic acid bacteria that grow under both aerobic and anaerobic conditions. In both examples, despite the heterogeneous flora of the finished product, a rather restricted group of microorganisms may develop and ultimately cause sensory changes in the product. Similar relationships exist for many other perishable foods.

It follows that since most classes of perishable foods constitute selective environments for rather restricted groups of microorganisms, the spoilage caused by the growth of these microorganisms manifests itself in a characteristic manner, i.e., normal spoilage pattern. For example, when pseudomonads and other closely related gram-negative psychrotrophic aerobic bacteria grow to large numbers on the surface of refrigerated fresh meat, poultry, and fish, sensory changes occur. The first manifestation of spoilage is development of off-odor. As growth proceeds, slime may develop and the off-odor may intensify. The normal spoilage pattern of a perishable food can be a safeguard, since under certain situations it warns the processor, preparer, or consumer that the food is no longer edible.

Changes in processing of perishable foods must take into account the effect these changes may have on the spoilage flora, and thus on the normal spoilage pattern of the food involved. If such changes tend to alter the normal patterns of spoilage the public health aspects must be taken into account. A classic example of this relates to the merchandising of smoked whitefish. For generations this product was merchandised under conditions where the fish was exposed to air. Spoilage was evidenced by the development of bacteria which produced off odors and slime that were readily recognized by the consumer and caused rejection of the product. Then it was discovered that the shelf-life of smoked fish could be significantly increased if the product was packed in an oxygen impermeable film. With extended storage of the product under these conditions, Clostridium *botulinum* (*C. botulinum* type E) was able to grow and produce toxin, just as it would have been able to do in the conventionally packaged product. However, under these storage conditions the aerobic bacteria producing off-odor and slime could not develop and the normal spoilage flora was now comprised of lactic acid bacteria that did not produce off-odors. This change in the normal spoilage pattern of the product reduced the probability that the consumer would reject a product that had been held in storage out of refrigeration for an extended period of time. This led to a multistate outbreak of type E botulism.

Approaches to microbiological control in

Three principal means have been used by regulatory agencies and food processors to control microorganisms in foods. These are

(1) education and training.

(2) inspection of facilities and operations.

(3) microbiological testing.

Although food handlers have the potential for contaminating foods with disease-producing microorganisms, i.e., staphylococci, salmonellae, and hepatitis virus, health examination of food handlers is a nonproductive approach to the control of foodborne illness. Specimens from food handlers have traditionally been examined only for a few microorganisms, and such tests do not always detect carriers. Screening tests cannot be made with sufficient frequency to be effective in detecting the carrier status in persons who are continually exposed to the risk of acquiring foodborne pathogens. Negative tests convey to food handlers, managers, and public health personnel the erroneous concept that the workers are free of infections and therefore incapable of transmitting foodborne pathogens to the foods they handle. Although direct transfer of pathogens from food handlers to food is a hazard, far more frequently improper food-handling practices create a hazard that is not circumvented by health examinations.

Education and Training Programs

These programs are directed primarily toward developing an understanding of the causes and consequences of microbial contamination and of measures to prevent contamination and subsequent growth. The extent of training required of personnel within processing plants and food service establishments depends upon the technical complexity of the food operation and the level of responsibility of the individuals being trained. In-depth training may be necessary for supervisory personnel, while for others training may relate only to specific aspects of a food operation. Although education and training are necessary parts of any food control program, standing alone they have certain limitations and shortcomings.

Personnel turnover in the food industry is both constant and rapid, and thus education of workers must be a continuing rather than a sporadic exercise. It is essential that supervisory personnel be properly trained with respect to the hazards associated with the operations for which they have responsibility.

Inspection of Facilities and Operations

Inspections of facilities and operations are commonly used to evaluate adherence to good handling practices. Resident inspectors observe all phases of processing from the live animal to the finished product.

Microbiological Testing

Samples of ingredients, materials obtained from selected points during the course of processing or handling, and finished products may be examined microbiologically to determine adherence to Good Manufacturing Practices. In some instances, foods are examined for a specific pathogen or its toxins, but more often examinations are made to detect organisms that are indicative of the possible presence of pathogens or spoilage or to detect presence of the specific spoilage organisms or their products. Microbiological testing is absolutely essential to the control of certain products, e.g., to assure that dried milk and eggs and confectionery products are free of a Salmonella hazard. Testing is essential to assure that critical raw materials are satisfactory for their intended use, e.g., to assure that the sugar used in canning meets established standards and to assure that critical products used in dried blends are free of Salmonella.

Microbiological testing has severe limitations as a control option. The most serious shortcoming is the constraint of time. Most microbiological test results are not available until several days after testing. Therefore, if finished product acceptability must be measured by microbiological testing, the product is held pending results. With perishable foods, this is generally not possible; with shelf stable foods, the warehousing of finished product increases costs. If in-line samples are collected and analyzed, the results are of retrospective value since the finished product has already been produced.

Practical: Food preservation

Objectives

Recognize food preservation related concepts.

List the different techniques of food preservation

Understand the principals of food preservation techniques

Food preservation prevents the growth of microorganisms (such as yeasts), or other microorganisms (although some methods work by introducing benign bacteria or fungi to the food), as well as slowing the oxidation of fats that cause rancidity. Food preservation may also include processes that inhibit visual deterioration, such as the enzymatic browning reaction in apples after they are cut during food preparation.

Traditional techniques

Curing

The earliest form of curing was dehydration or drying, used as early as 12,000 BC. Smoking and salting techniques improve on the drying process and add antimicrobial agents that aid in preservation. Smoke deposits a number of pyrolysis products onto the food, including the phenols syringol, guaiacol and catechol. Salt accelerates the drying process using osmosis and also inhibits the growth of several common strains of bacteria. More recently nitrites have been used to cure meat, contributing a characteristic pink color.

Cooling

preserves food by slowing down the growth and reproduction of microorganisms and the action of enzymes that causes the food to rot. The introduction of commercial and domestic refrigerators drastically improved the diets of many in the Western world by allowing food such as fresh fruit, salads and dairy products to be stored safely for longer periods, particularly during warm weather.

Before the era of mechanical refrigeration, cooling for food storage occurred in the forms of root cellars and iceboxes. Rural people often did their own ice cutting, whereas town and city dwellers often relied on the ice trade. Today, root cellaring remains popular among people who value various goals, including local food, heirloom crops, traditional home cooking techniques, family farming, frugality, self-sufficiency, organic farming, and others.

Freezing

is also one of the most commonly used processes, both commercially and domestically, for preserving a very wide range of foods, including prepared foods that would not have required freezing in their unprepared state. For example, potato waffles are stored in the freezer, but potatoes themselves require only a cool dark place to ensure many months' storage. Cold stores provide large-volume, long-term storage for strategic food stocks held in case of national emergency in many countries.

Boiling

Boiling liquid food items can kill any existing microbes. Milk and water are often boiled to kill any harmful microbes that may be present in them.

Heating

Heating to temperatures which are sufficient to kill microorganisms inside the food is a method used with perpetual stews. Milk is also boiled before storing to kill many microorganisms.

Sugaring

The earliest cultures have used sugar as a preservative, and it was commonplace to store fruit in honey. Sugar tends to draw water from the microbes (plasmolysis). This process leaves the microbial cells dehydrated, thus killing them. In this way, the food will remain safe from microbial spoilage."[5] Sugar is used to preserve fruits, either in an antimicrobial syrup with fruit such as apples, pears, peaches, apricots, and plums, or in crystallized form where the preserved material is cooked in sugar to the point of crystallization and the resultant product is then stored dry. This method is used for the skins of citrus fruit (candied peel), angelica, and ginger. Also, sugaring can be used in the production of jam and jelly.

Pickling

Pickling is a method of preserving food in an edible, antimicrobial liquid. Pickling can be broadly classified into two categories: chemical pickling and fermentation pickling.

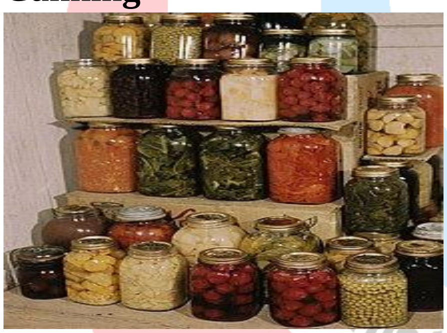
In chemical pickling, the food is placed in an edible liquid that inhibits or kills bacteria and other microorganisms. Typical pickling agents include brine (high in salt), vinegar, alcohol, and vegetable oil. Many chemical pickling processes also involve heating or boiling so that the food being preserved becomes saturated with the pickling agent. Common chemically pickled foods include cucumbers, peppers, corned beef, herring, and eggs, as well as mixed vegetables such as piccalilli.

In fermentation pickling, bacteria in the liquid produce organic acids as preservation agents, typically by a process that produces lactic acid through the presence of lactobacillales. Fermented pickles include sauerkraut, nukazuke, kimchi, and surströmming.

Lye

Sodium hydroxide (lye) makes food too alkaline for bacterial growth. Lye will saponify fats in the food, which will change its flavor and texture. Lutefisk uses lye in its preparation, as do some olive recipes. Modern recipes for century eggs also call for lye.

Canning



Canning involves cooking food, sealing it in sterilized cans or jars, and boiling the containers to kill or weaken any remaining bacteria as a form of sterilization. It was invented by the French confectioner Nicolas Appert. By 1806, this process was used by the French Navy to preserve meat, fruit, vegetables, and even milk. Although Appert had discovered a new way of preservation, it wasn't understood until 1864 when Louis Pasteur found the relationship between microorganisms, food spoilage, and illness.

Foods have varying degrees of natural protection against spoilage and may require that the final step occur in a pressure cooker. High-acid fruits like strawberries require no preservatives to can and only a short boiling cycle, whereas marginal vegetables such as carrots require longer boiling and addition of other acidic elements. Low-acid foods, such as vegetables and meats, require pressure canning. Food preserved by canning or bottling is at immediate risk of spoilage once the can or bottle has been opened.

Lack of quality control in the canning process may allow ingress of water or micro-organisms. Most such failures are rapidly detected as decomposition within the can causes gas production and the can will swell or burst. However, there have been examples of poor manufacture (under processing) and poor hygiene allowing contamination of canned food by the obligate anaerobe *Clostridium botulinum*, which produces an acute toxin within the food, leading to severe illness or death. This organism produces no gas or obvious taste and remains undetected by taste or smell. Its toxin is denatured by cooking, however. Cooked mushrooms, handled poorly and then canned, can support the growth of Staphylococcus aureus, which produces a toxin that is not destroyed by canning or subsequent reheating.

Jellying

Food may be preserved by cooking in a material that solidifies to form a gel. Such materials include gelatin, agar, maize flour, and arrowroot flour. Some foods naturally form a protein gel when cooked, such as eels and elvers, and sipunculid worms, which are a delicacy in Xiamen, in the Fujian province of the People's Republic of China. Jellied eels are a delicacy in the East End of London, where they are eaten with mashed potatoes. Potted meats in aspic (a gel made from gelatin and clarified meat broth) were a common way of serving meat off-cuts in the UK until the 1950s. Many jugged meats are also jellied.

A traditional British way of preserving meat (particularly shrimp) is by setting it in a pot and sealing it with a layer of fat. Also common is potted chicken liver; jellying is one of the steps in producing traditional pâtés.

Jugging

Meat can be preserved by jugging. Jugging is the process of stewing the meat (commonly game or fish) in a covered earthenware jug or casserole. The animal to be jugged is usually cut into pieces, placed into a tightly-sealed jug with brine or gravy, and stewed. Red wine and/or the animal's own blood is sometimes added to the cooking liquid. Jugging was a popular method of preserving meat up until the middle of the 20th century.

Burial

Burial of food can preserve it due to a variety of factors: lack of light, lack of oxygen, cool temperatures, pH level, or desiccants in the soil. Burial may be combined with other methods such as salting or fermentation. Most foods can be preserved in soil that is very dry and salty (thus a desiccant) such as sand, or soil that is frozen.

Many root vegetables are very resistant to spoilage and require no other preservation than storage in cool dark conditions, for example by burial in the ground, such as in a storage clamp. Century eggs are traditionally created by placing eggs in alkaline mud (or other alkaline substance), resulting in their "inorganic" fermentation through raised pH instead of spoiling. The fermentation preserves them and breaks down some of the complex, less flavorful proteins and fats into simpler, more flavorful ones. Cabbage was traditionally buried during Autumn in northern US farms for preservation. Some methods keep it crispy while other methods produce sauerkraut. A similar process is used in the traditional production of kimchi. Sometimes meat is buried under conditions that cause preservation. If buried on hot coals or ashes, the heat can kill pathogens, the dry ash can desiccate, and the earth can block oxygen and further contamination. If buried where the earth is very cold, the earth acts like a refrigerator. Before burial, meat (pig/boar) can be fatted. The tallow of the animal is heated and poured over meat in a barrel. Once the fat hardens the barrel is sealed and buried in a cold cellar or ground.

Fermentation

Some foods, such as many cheeses, wines, and beers, use specific micro-organisms that combat spoilage from other less-benign organisms. These micro-organisms keep pathogens in check by creating an environment toxic for themselves and other micro-organisms by producing acid or alcohol. Methods of fermentation include, but are not limited to, starter micro-organisms, salt, hops, controlled (usually cool) temperatures and controlled (usually low) levels of oxygen. These methods are used to create the specific controlled conditions that will support the desirable organisms that produce food fit for human consumption.

Fermentation is the microbial conversion of starch and sugars into alcohol. Not only can fermentation produce alcohol, but it can also be a valuable preservation technique. Fermentation can also make foods more nutritious and palatable. For example, drinking water in the Middle Ages was dangerous because it often contained pathogens that could spread disease. When the water is made into beer, the boiling during the brewing process kills any bacteria in the water that could make people sick.

Additionally, the water now has the nutrients from the barley and other ingredients, and the microorganisms can also produce vitamins as they ferment.

Modern industrial techniques

Techniques of food preservation were developed in research laboratories for commercial applications.

Pasteurization

Pasteurization is a process for preservation of liquid food. It was originally applied to combat the souring of young local wines. Today, the process is mainly applied to dairy products. In this method, milk is heated at about 70 °C (158 °F) for 15–30 seconds to kill the bacteria present in it and cooling it quickly to 10 °C (50 °F) to prevent the remaining bacteria from growing. The milk is then stored in sterilized bottles or pouches in cold places.

Vacuum packing

Vacuum-packing stores food in a vacuum environment, usually in an air-tight bag or bottle. The vacuum environment strips bacteria of oxygen needed for survival. Vacuum-packing is commonly used for storing nuts to reduce loss of flavor from oxidization. A major drawback to vacuum packaging, at the consumer level, is that vacuum sealing can deform contents and rob certain foods, such as cheese, of its flavor.

Artificial food additives

Preservative food additives can be *antimicrobial*—which inhibit the growth of bacteria or fungi, including mold—or *antioxidant*, such as oxygen absorbers, which inhibit the oxidation of food constituents. Common antimicrobial preservatives include calcium propionate, sodium nitrate, sodium nitrite, sulfites (sulfur dioxide, sodium bisulfite, potassium hydrogen sulfite, etc.). Antioxidants include butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT). Other preservatives include formaldehyde (usually in solution), glutaraldehyde (insecticide), ethanol, and methylchloroisothiazolinone.

Irradiation

Irradiation of foodis the exposure of food to ionizing radiation. Multiple types of ionizing radiation can be used, including beta particles (high-energy electrons) and gamma rays (emitted from radioactive sources such as cobalt-60 or cesium-137). Irradiation can kill bacteria, molds, and insect pests, reduce the ripening and spoiling of fruits, and at higher doses induce sterility. The technology may be compared to pasteurization; it is sometimes called "cold pasteurization", as the product is not heated. Irradiation may allow lower-quality or contaminated foods to be rendered marketable.

National and international expert bodies have declared food irradiation as "wholesome"; organizations of the United Nations, such as the World Health Organization and Food and Agriculture Organization, endorse food irradiation. Consumers may have a negative view of irradiated food based on the misconception that such food is radioactive;[12] in fact, irradiated food does not and cannot become radioactive. Activists have also opposed food irradiation for other reasons, for example, arguing that irradiation can be used to sterilize contaminated food without resolving the underlying cause of the contamination.

Pulsed electric field electroporation

Pulsed electric field (PEF) electroporation is a method for processing cells by means of brief pulses of a strong electric field. PEF holds potential as a type of low-temperature alternative pasteurization process for sterilizing food products. In PEF processing, a substance is placed between two electrodes, then the pulsed electric field is applied. The electric field enlarges the pores of the cell membranes, which kills the cells and releases their contents. PEF for food processing is a developing technology still being researched. There have been limited industrial applications of PEF processing for the pasteurization of fruit juices. For cell disintegration purposes especially potato processors show great interest in PEF technology as an efficient alternative for their preheaters.

Modified atmosphere

Modifying atmosphere is a way to preserve food by operating on the atmosphere around it. Salad crops that are notoriously difficult to preserve are now being packaged in sealed bags with an atmosphere modified to reduce the oxygen (O_2) concentration and increase the carbon dioxide (CO_2) concentration. There is concern that, although salad vegetables retain their appearance and texture in such conditions, this method of preservation may not retain nutrients, especially vitamins. There are two methods for preserving grains with carbon dioxide. One method is placing a block of dry ice in the bottom and filling the can with the grain. Another method is purging the container from the bottom by gaseous carbon dioxide from a cylinder or bulk supply vessel.

Carbon dioxide prevents insects and, depending on concentration, mold and oxidation from damaging the grain. Grain stored in this way can remain edible for approximately five years.

Nitrogen gas (N_2) at concentrations of 98% or higher is also used effectively to kill insects in the grain through hypoxia. However, carbon dioxide has an advantage in this respect, as it kills organisms through hypercarbia and hypoxia (depending on concentration), but it requires concentrations of above 35%,or so. This makes carbon dioxide preferable for fumigation in situations where a hermetic seal cannot be maintained.

Controlled Atmospheric Storage (CA): "CA storage is a non-chemical process. Oxygen levels in the sealed rooms are reduced, usually by the infusion of nitrogen gas, from the approximate 21 percent in the air we breathe to 1 percent or 2 percent. Temperatures are kept at a constant 0–2 °C (32–36 °F). Humidity is maintained at 95 percent and carbon dioxide levels are also controlled. Exact conditions in the rooms are set according to the apple variety.

Air-tight storage of grains (sometimes called hermetic storage) relies on the respiration of grain, insects, and fungi that can modify the enclosed atmosphere sufficiently to control insect pests. This is a method of great antiquity, as well as having modern equivalents. The success of the method relies on having the correct mix of sealing, grain moisture, and temperature.

A patented process uses fuel cells to exhaust and automatically maintain the exhaustion of oxygen in a shipping container, containing, for example, fresh fish.

Nonthermal plasma

This process subjects the surface of food to a "flame" of ionized gas molecules, such as helium or nitrogen. This causes micro-organisms to die off on the surface.

High-pressure food preservation

High-pressure food preservation or pascalization refers to the use of a food preservation technique that makes use of high pressure. "Pressed inside a vessel exerting 70,000 pounds per square inch or more, food can be processed so that it retains its fresh appearance, flavor, texture and nutrients while disabling harmful microorganisms and slowing spoilage." By 2005, the process was being used for products ranging from orange juice to guacamole to deli meats and widely sold.

Biopreservation is the use of natural or controlled microbiota or antimicrobials as a way of preserving food and extending its shelf life.[27] Beneficial bacteria or the fermentation products produced by these bacteria are used in biopreservation to control spoilage and render pathogens inactive in food.[28] It is a benign ecological approach which is gaining increasing attention.

Of special interest are lactic acid bacteria (LAB). Lactic acid bacteria have antagonistic properties that make them particularly useful as biopreservatives. When LABs compete for nutrients, their metabolites often include active antimicrobials such as lactic acid, acetic acid, hydrogen peroxide, and peptide bacteriocins. Some LABs produce the antimicrobial nisin, which is a particularly effective preservative.

These days, LAB bacteriocins are used as an integral part of hurdle technology. Using them in combination with other preservative techniques can effectively control spoilage bacteria and other pathogens, and can inhibit the activities of a wide spectrum of organisms, including inherently resistant Gram-negative bacteria.

Hurdle technology

Hurdle technology is a method of ensuring that pathogens in food products can be eliminated or controlled by combining more than one approach. These approaches can be thought of as "hurdles" the pathogen has to overcome if it is to remain active in the food. The right combination of hurdles can ensure all pathogens are eliminated or rendered harmless in the final product.

Hurdle technology has been defined by Leistner (2000) as an intelligent combination of hurdles that secures the microbial safety and stability as well as the organoleptic and nutritional quality and the economic viability of food products.[32] The organoleptic quality of the food refers to its sensory properties, that is its look, taste, smell, and texture.

Examples of hurdles in a food system are high temperature during processing, low temperature during storage, increasing the acidity, lowering the water activity or redox potential, and the presence of preservatives or biopreservatives. According to the type of pathogens and how risky they are, the intensity of the hurdles can be adjusted individually to meet consumer preferences in an economical way, without sacrificing the safety of the product.

Chapter 5: Interaction between Malnutrition and Infection

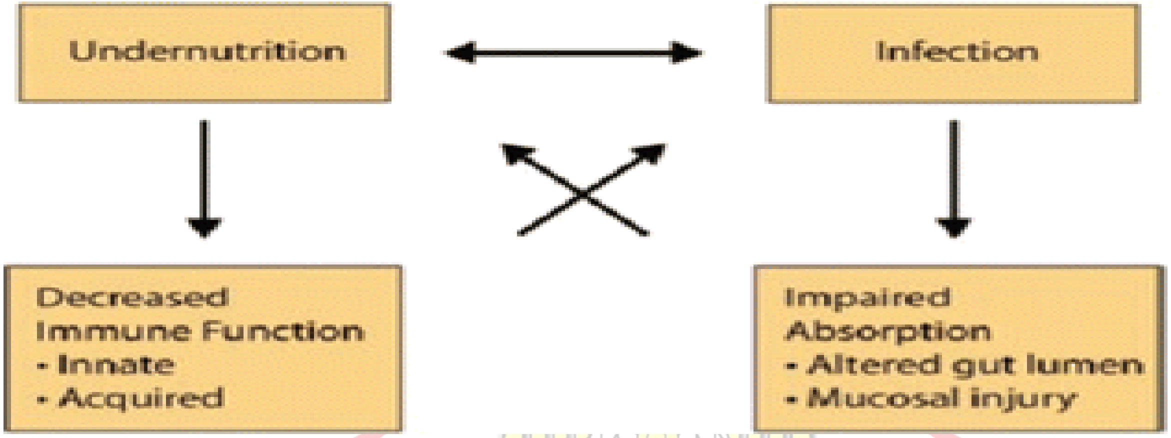
Objectives

Understand the cycle of malnutrition and infection.
List the commonest causes of malnutrition.

Overview of malnutrition and related concepts

Malnutrition is a condition that results from eating a diet in which one or more nutrients are either not enough or are too much such that the diet causes health problems. It may involve calories, protein, carbohydrates, vitamins or minerals. Not enough nutrients is called undernutrition or undernourishment while too much is called overnutrition. Malnutrition is often used to specifically refer to undernutrition where an individual is not getting enough calories, protein, or micronutrients. If undernutrition occurs during pregnancy, or before two years of age, it may result in permanent problems with physical and mental development. Extreme undernourishment, known as starvation, may have symptoms that include: a short height, thin body, very poor energy levels, and swollen legs and abdomen.

Malnutrition is the primary cause of immunodeficiency worldwide, with infants, children, adolescents, and the elderly most affected. There is a strong relationship between malnutrition and infection and infant mortality, because poor nutrition leaves children underweight, weakened, and vulnerable to infections, primarily because of epithelial integrity and inflammation. Five infectious diseases—pneumonia, diarrhea, malaria, measles, and AIDS—account for more than one-half of all deaths in children aged <5 years. In our understanding of this interaction between infection and malnutrition, it is important to remember that a decreased immune function is not always a defective one, and many indicators of nutritional status are not reliable during infection.

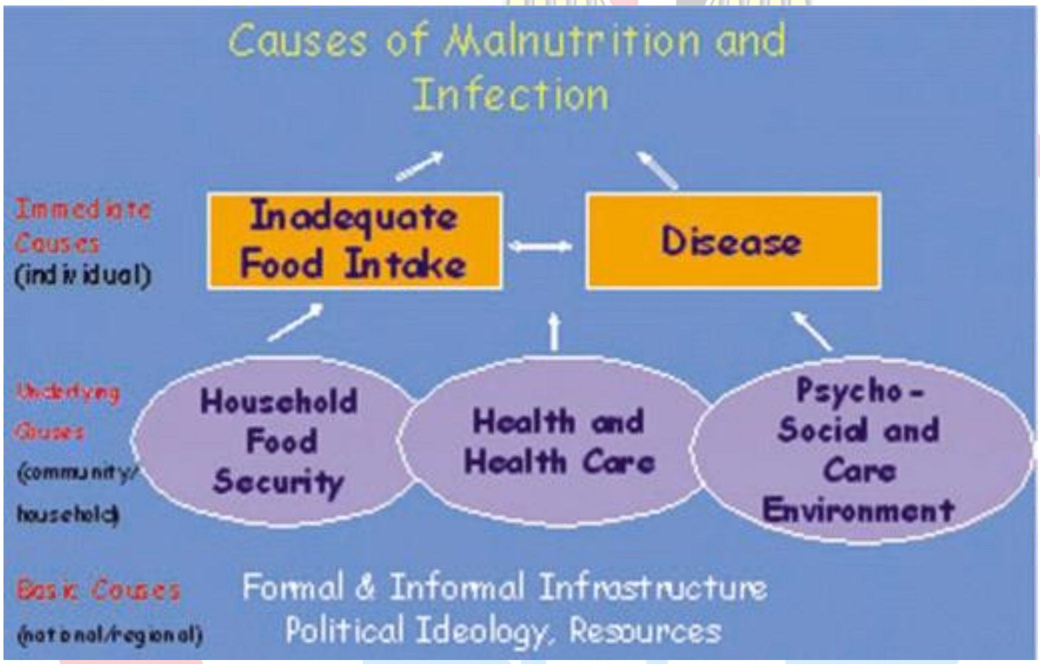


The Cycle of Malnutrition and Infection

Malnutrition can make a person more susceptible to infection, and infection also contributes to malnutrition, which causes a vicious cycle. An inadequate dietary intake leads to weight loss, lowered immunity, mucosal damage, invasion by pathogens, and impaired growth and development in children. A sick person's nutrition is further aggravated by diarrhea, malabsorption, loss of appetite, diversion of nutrients for the immune response, and urinary nitrogen loss, all of which lead to nutrient losses and further damage to defense mechanisms. These, in turn, cause reduced dietary intake. In addition, fever increases both energy and micronutrient requirements. Malaria and influenza, for example, have mortality rates proportionate to the degree of malnutrition

The causes of malnutrition

The factors responsible are household food availability, personal health, health services, and the psychosocial care environment. The existing primary health care infrastructure includes the types of services provided and the accessibility of health care (distance and affordability). Underlying the problem of malnutrition and disease is inadequate household food security, which is defined as “access by all members at all times to enough food for an active, healthy life,” not merely as adequate food for survival. Access to health services and environmental health conditions relate to essential drugs and immunizations, safe water, sanitation, and housing. Insufficient or delayed treatment also prolongs disease occurrence and severity.



Many of the basic causes of malnutrition also emerge at the national and international levels and relate to the availability and control of food. The political ideology of the ruling government and its commitment to preventing infectious disease and malnutrition affects the health of its entire population. Food supply, underlying health, and health care interact in important ways, and their combined effect is synergistic. The underlying causes may also change with the seasons. Rural households, for example, may experience an annual hunger season. Diarrheal diseases and malaria are more prevalent during rainy seasons, and respiratory tract infections are more prevalent during cold weather.

Practical: The role of Vitamins in Immune Function

Objectives

Discuss the role of vitamins in immune function.

Discuss the role of minerals in immune function.

Recognize the interaction between vitamins and infection.

Worldwide, 2 billion people are affected by micronutrient deficiencies, including vitamins A, C, and E and minerals zinc, iron, and iodine. The effects are poor growth, impaired intellect, and increased mortality and susceptibility to infection. Micronutrients have a relationship to antibody formation and the development of the immune system. These ill effects are preventable by supplements, fortification, and diet change.

Vitamin A maintains the integrity of the epithelium in the respiratory and gastrointestinal tracts. Vitamin A deficiency increases the risk of diarrhea, measles, and overall mortality.

Vitamin A deficiency and measles, which is estimated to kill 2 million children per year, are closely linked. Measles in a child is more likely to exacerbate any existing nutritional deficiency, and children who are already deficient in vitamin A are at much greater risk of dying from measles. Post measles diarrhea is particularly difficult to treat and has a very high mortality. Vitamin A deficiency increases the risk of developing respiratory disease and chronic ear infections . Vitamin A supplementation sustains gut integrity, lowers the incidence of respiratory tract infections, reduces mortality from diarrhea, and enhances immunity. Measles also depletes the body's supply of vitamin A. Thus, vaccination against measles often includes a high dose of vitamin A.

Vitamin E is an antioxidant that scavenges free radicals. Vitamin E supplementation has been shown to improve immune function in the elderly, with delayed hypersensitivity skin response and antibody production after vaccination. Vitamin E increases both cell-dividing and interleukin-producing capacities of naive T cells but not of memory T cells. This enhancement of immune function is associated with significant improvement in resistance to influenza virus infection in aged mice and a reduced risk of acquiring upper respiratory infections in nursing home residents.

Vitamin C Major dietary sources of vitamin C are vegetables and fruit. vitamin C is a water-soluble substance found in body fluids rather than in cellular lipids and membranes. Vitamin C acts as a major antioxidant in the aqueous phase and also reinforces the effects of other antioxidants, such as vitamin E, by regenerating their active forms after they have reacted with free radicals. Its Antioxidant function protects phagocytes. the Deficiency of vitamin C lowers phagocytes activity, tumor resistance, graft rejection, and slows wound repair.

Vitamin D is a hormone, and its precursor can be synthesised in the skin by sunlight exposure. The precursor is first metabolised in the liver and then in the kidney to its biologically active form of vitamin D. However, latitude, season, skin pigmentation, aging and sunscreen use all may decrease cutaneous vitamin D production. Dietary sources of vitamin D precursor are oily fish, cod-liver oil and egg yolk. It is estimated that one billion people worldwide have vitamin D deficiency or insufficiency. Vitamin D is an important immune system regulator. Receptors for this vitamin occur in most cells of the body, including immune cells. Liver cells, kidney cells and macrophages have the enzymatic capacity to make the biologically active form of vitamin D. Vitamin D supplements may offer a cheap and effective immune system boost against tuberculosis. Vitamin D was used to treat tuberculosis in the preantibiotic era.

Vitamin B is widely distributed in foods, with rich sources including fowl, fish, liver, cereals and pulses. deficiencies in vitamin B,impair immune function. vitamin B is essential for a wide variety of reactions necessary for the synthesis and metabolism of amino acids (the building blocks of protein), and there is a need for increased protein synthesis during the immune response. deficiency of vitamin B impairs both antibody production and T cell activity. Lymphocyte growth and maturation are also altered, and natural killer cell activity is decreased.

Zinc is a trace mineral that is essential for all species and is required for the activities of >300 enzymes, carbohydrate and energy metabolism, protein synthesis and degradation, nucleic acid production, heme biosynthesis, and carbon dioxide transport. It is a cofactor in the formation of enzymes and nucleic acids and plays a critical role in the structure of cell membranes and in the function of immune cells. Zinc deficiency reduces nonspecific immunity, including neutrophil and natural killer cell function and complement activity; reduces numbers of T and B lymphocytes; and suppresses delayed hypersensitivity, cytotoxic activity, and antibody production. Inadequate zinc supply prevents normal release of vitamin A from the liver; clinically, it is associated with growth retardation, malabsorption syndromes, fetal loss, neonatal death, and congenital abnormalities. Low blood zinc concentrations have also been found in patients with tuberculosis, Crohn disease, diarrheal disease, and pneumonia. Zinc deficiency is associated with abnormal pregnancy outcomes and conditions of relative immunocompromise, including alcoholism, kidney disease, burns, inflammatory bowel disease, and HIV infection. Many drugs, including corticosteroids, also cause excessive excretion.

Selenium Dietary selenium occurs in protein-rich foods such as meat, fish, nuts and seeds. Selenium is essential for an optimal immune response and influences both the innate and acquired immune system. It has a key role in the redox balance, including the protection against DNA damage. Selenium is also an important cofactor of a group of enzymes that contribute to the protection of cells from oxidative damage. Because phagocytes generate large amounts of reactive oxygen species, selenium may be a factor in protecting phagocytes from an excess of such oxidants. However, evaluation of the direct effects of selenium is difficult because of interactions between selenium and another antioxidant, vitamin E.

Other minerals

In addition to zinc and selenium other minerals and trace elements are important for the normal functioning of the immune system. These include iron, copper, magnesium and manganese. Iron is an example of a nutrient, which is not only required for an adequate immune response but also for an optimal growth of pathogens. As long as iron deficiency does not impair immune function, iron supplementation may only benefit pathogenic bacteria. Therefore, supplementation of this trace element has to be considered on an individual basis

Carotenoids are yellow, orange and red compounds found in fruits and vegetables. Examples are β-carotene, which is widely distributed in plants, and lycopene, a carotenoid found in tomatoes. Like vitamins C and E, carotenoids are antioxidants, and, additionally, β-carotene is a precursor of vitamin A. diets rich in carotenoids reduce the risk of respiratory infections. A high dietary intake of carotenoids is further inversely associated with inflammatory markers suggesting that carotenoids possess an anti-inflammatory. β-carotene supplementation may be beneficial for individuals with a compromised immune system.

Flavonoids occur in all plant foods and include several thousands of compounds with well-defined chemical structures. They are responsible for the red and blue colours in plant foods as well as for some typical flavours of plants. flavonoids have immunosuppressive effects. a subgroup of flavonoids, the anthocyanins, possesses anti-inflammatory activities at dietary relevant doses.

Effects of deficiency or insufficiency in micronutrients or phytochemicals on the immune response

Micronutrient	Effects of deficiency or insufficiency
Vitamin A	Loss of mucosal epithelial barrier function Impaired neutrophil and macrophage function Decreased natural killer cell number and lytic activity Diminished antibody response
Vitamin D	Decreased production of antibacterial peptides
Vitamin E	Impaired B- and T-cell-mediated immunity Increased oxidative damage in immune cell membranes
Vitamin B_6	Decreased lymphocyte responsiveness Impaired antibody production
Vitamin C	Decreased resistance to infection Impaired proliferation of T cells
Zinc	Impaired T cell development Impaired lymphocyte responsiveness Decreased resistance to infection
Selenium	Increased oxidative damage in immune cell membranes Decreased cytokine production Decreased resistance to viruses Decreased antibody production
Carotenoids	Decreased natural killer cell activity Decreased production of cytokines Impaired function of phagocytes
Flavonoids	Immunomodulating effects

Chapter 6: Nutrition and Immunity

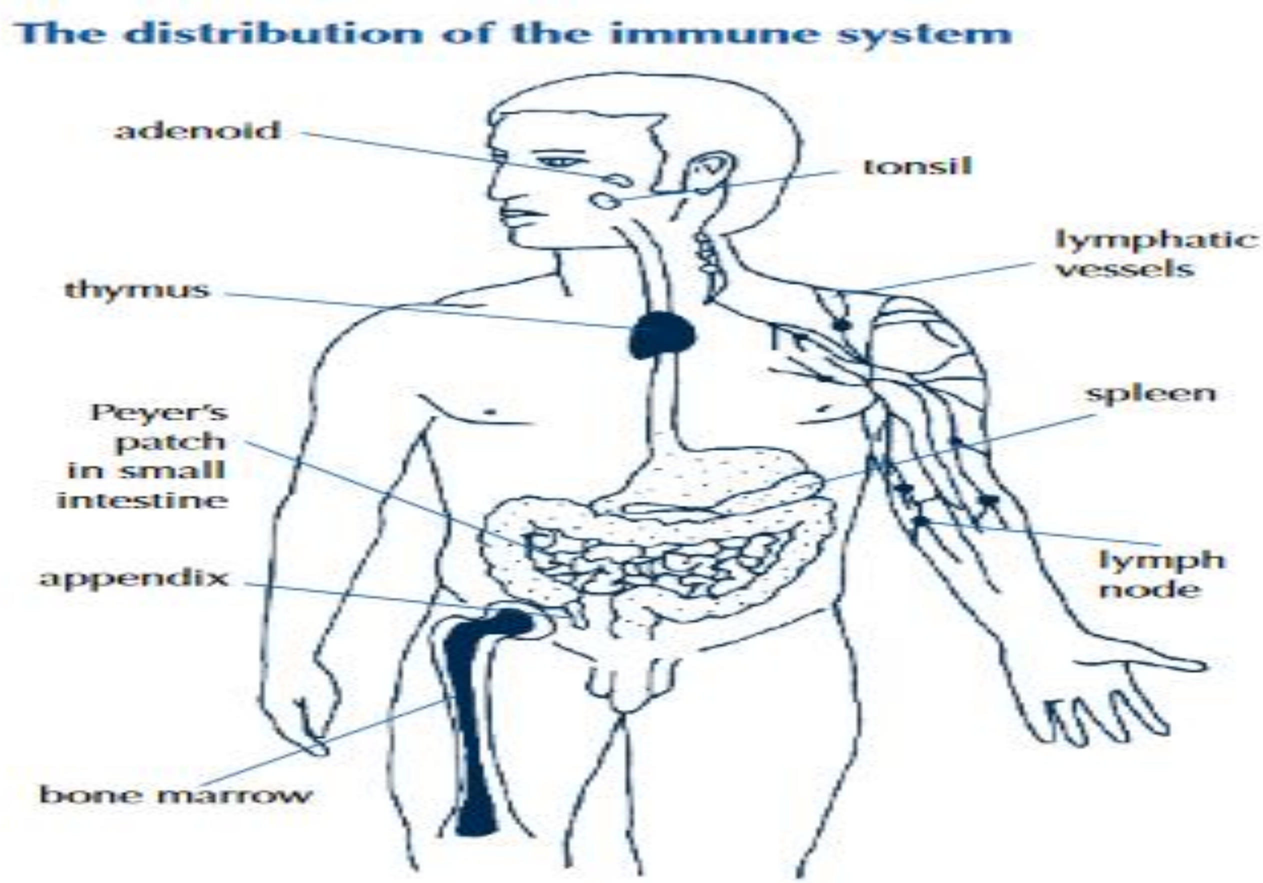
Objectives

Explain how a deficiency, excess or imbalance of nutrients may affect immunity.

Explain how inflammation is involved in chronic diseases: obesity, cardiovascular disease and type 2 diabetes.

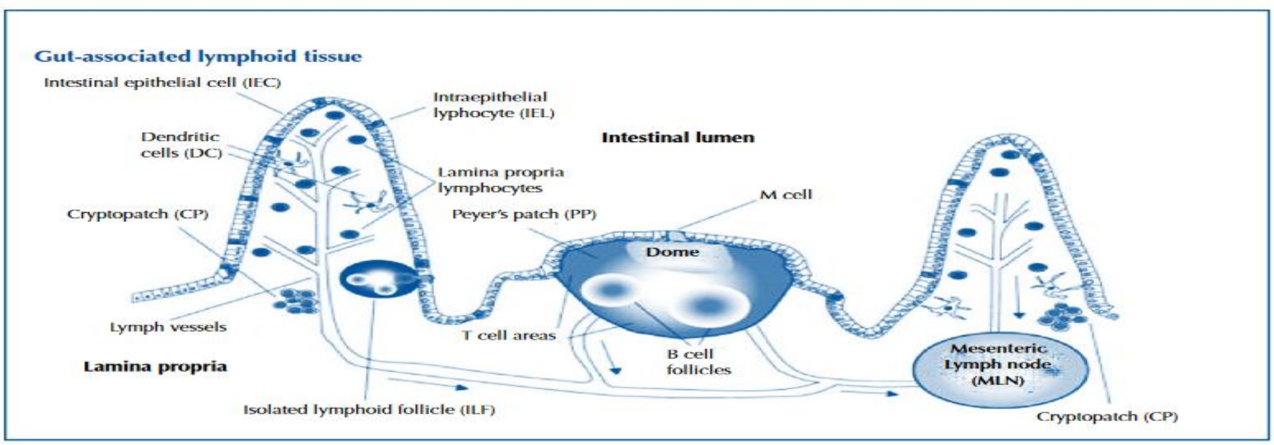
Overview of the immune system and related concepts

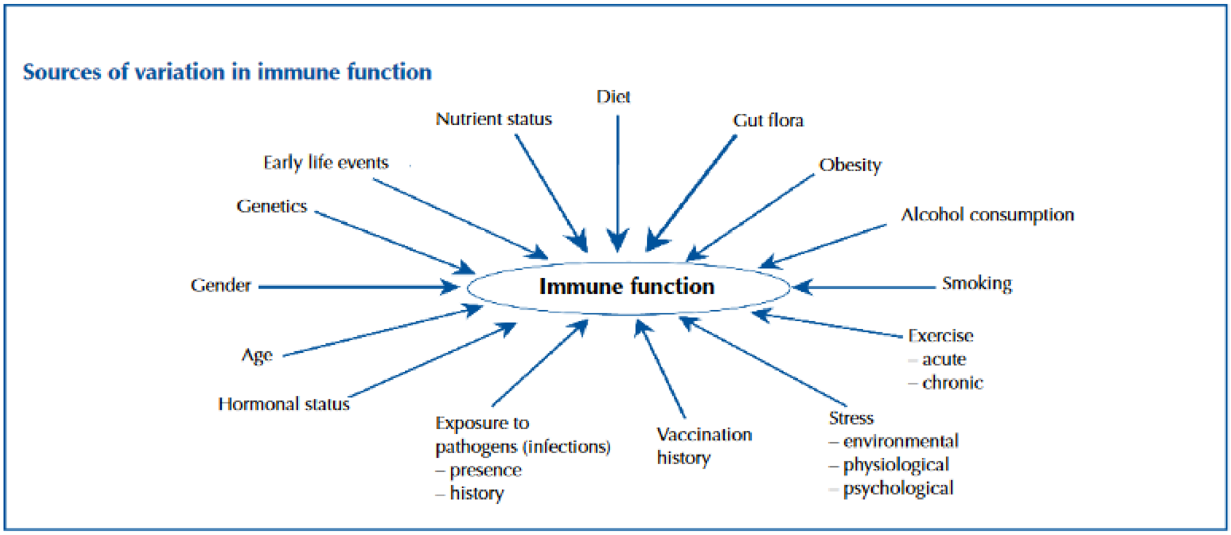
The human body has an intricate system of defense mechanisms, which protects it against potentially harmful foreign agents. This complex system of effector molecules, cells and tissues is widely dispersed throughout the body. The body has various nonspecific defense mechanisms like skin and mucous secretions. Any organism that breaks through this surface barrier encounters two further levels of defense, the innate and the acquired immune responses.



The immune system of the gut

The gut immune system or the gut-associated lymphoid tissue (GALT) is an important part of the total immunological capacity of an individual. The GALT prevents the passage of bacteria and food antigens from the gastrointestinal lumen through the epithelium of the intestinal mucosa. It does allow, however, the translocation of minute amounts of viable and dead bacteria as important immunological information to the systemic immune system. Immune cells of the gut are organized in different compartments, such as lymph nodes, lymph follicles and Peyer's patches. Single immune cells are distributed within the intestinal mucosa and between epithelial cells. To cope with various challenges, the GALT has developed at least two strategies. First, it provides for immune exclusion by secreting antibodies to inhibit the colonization of disease-causing bacteria and to prevent mucosal infections. Second, the GALT possesses mechanisms to avoid overreaction to innocuous substances from occurring on mucosal surfaces. The latter phenomenon is called oral tolerance and largely explains why most people show no adverse immune reactions to foods. However, in some individuals the immune system initiates an inappropriate and exaggerated immune response towards food constituents, which is known as "food allergy".



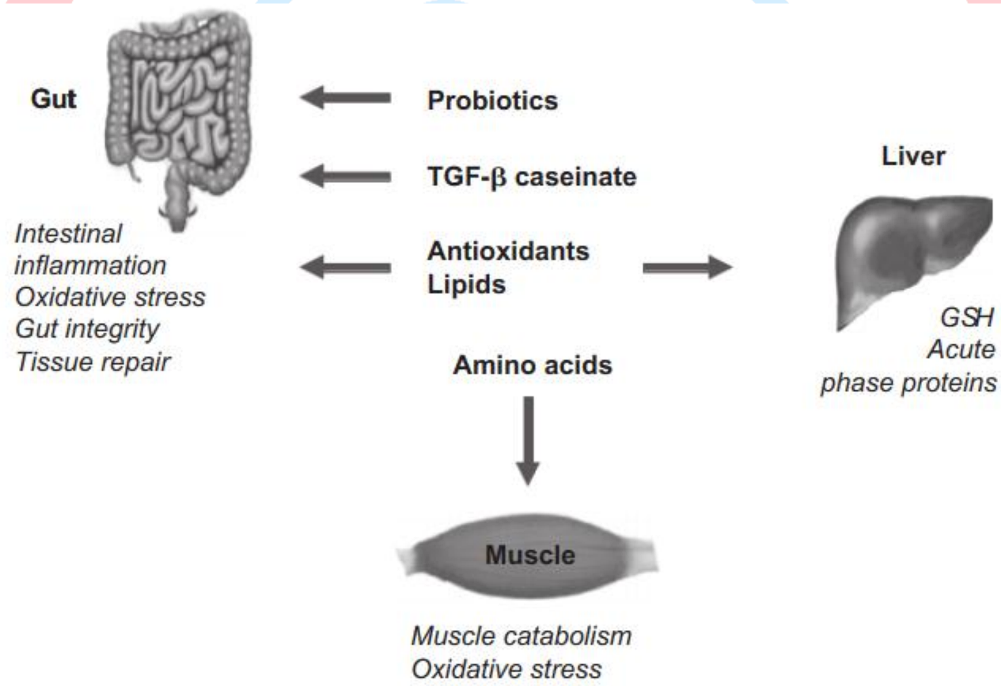


Dietary factors which alter the immune response:

The recommended daily amount (RDA) for a nutrient indicates the average daily dietary intake level considered sufficient to meet the requirements of nearly all (97–98%) healthy individuals. However, for most nutrients their role in supporting an optimal immune response has not been considered when defining the RDA. Therefore, the RDA for a specific nutrient may not be at a level that allows the best immune response. High and low energy intakes adversely affect the immune response. It is well known that severe malnutrition, especially wasting malnutrition in children, leads to impairments in immune function. Such malnutrition, which is primarily a problem in developing countries, substantially increases the risk of childhood mortality from infection. Protein-energy malnutrition is often accompanied by deficiencies of micronutrients such as vitamin A, vitamin E, vitamin B6, vitamin C, folate, zinc, iron, copper and selenium. Most host defense mechanisms are impaired in malnutrition, even if the nutritional deficiency is only moderate in severity. The rapidly proliferating T cells responding to pathogens are especially affected, resulting in a decrease in their numbers. Severe and chronic malnutrition leads to atrophy of the thymus and other lymphoid organs. Both obesity and its treatment seem to have clear but not

Inflammation is a basic process whereby tissues of the body respond to injury.Inflammation has been described as purposeful, timely, powerful and, as aconsequence, also as dangerous, if resolution is not initiated. The normal outcome of the acute inflammatory programme is successful resolution and repair of tissue damage, rather than persistence of the inflammatory response. Natural resolution of inflammation is a highly complex, multifactorial and tightly controlled process driven by removal of the initial stimulus, decrease in pro-inflammatory mediators (mainly cytokines, chemokines), elimination of damaged and inflammatory cells, and promotion of repair. Although inflammation is essential for tissue homeostasis, prolonged inflammation is a hallmark of many chronic diseases such as inflammatory bowel disease and auto-immunity. Moreover, chronic inflammation has been shown to be implicated in critical conditions such as atherosclerosis, arthritis, cancer and asthma all leading to tissue destruction, fibrosis and impairment or loss of organ function.

inflammation can be managed by nutritional means. Probiotics, antioxidants and lipids influence intestinal inflammation, gut integrity, tissue repair and oxidative stress. Antioxidants and lipids also modulate acute phase proteins and the glutathione. Moreover, free amino acids feed into muscle catabolism and can influence the oxidative stress in the muscle.



Managing inflammation by nutritional means.

Examples of diseases with an inflammatory background

Diseases	Characteristics
Obesity/metabolic syndrome	Production of pro-inflammatory cytokines, such as TNF-α and IL-6, by adipose tissue
Atherosclerosis	Inflammatory disease of the arteries resulting in the arterial deposition of plaques
Asthma	Chronic inflammation of the lung mucosa accompanied by a contraction of the smooth muscle layer in the bronchi
Contact dermatitis and psoriasis	Inflammatory skin diseases characterised by T cell differentiation towards T_h2 phenotype (contact dermatitis) or failure of regulatory processes during inflammation (psoriasis)
Rheumatoid arthritis	Autoimmune disease characterised by a chronic inflammation of the synovial membrane in the joints
Crohn's disease and ulcerative colitis	Inflammatory bowel diseases characterised by acute and chronic inflammation of the gut

Cardiovascular disease

Numerous dietary factors have been correlated with increases or decreases in the risk of heart disease. Atherosclerosis is a major cause of mortality from heart disease. It is an inflammatory disease of the arteries resulting in the deposition of arterial plaques. The immune system is involved in the pathogenesis of atherosclerosis through interaction between its white blood cells and cells of the arterial wall. Dietary factors influence the immunological processes underlying the pathogenesis of atherosclerosis. The risk of heart disease can be reduced by increased dietary antioxidants (vitamin C, vitamin E).

Obesity:

Chronic inflammation that lasts for a long time and is characterized by the presence of lymphocytes and macrophages and the proliferation of blood vessels and connective tissue is considered a characteristic feature of metabolic syndrome which is characterized by secretion of inflammatory adipokines usually from adipose tissue, such as leptin, interleukin (IL-6), tumor necrosis factor-α (TNF-α) and resistin . Obesity, which is a feature of metabolic syndrome, was associated with chronic inflammation in obese subjects.

Type 2 diabetes:

Low-grade inflammation is a common feature in subjects with type 2 diabetes (T2D). Heart disease, the metabolic syndrome and T2D all have in common the increased concentration of circulatory cytokines as a result of inflammation. Inflammatory cytokines are produced by different cell types and secreted into the circulation, where they regulate different tissues through their local, central and peripheral actions. C-reactive protein (CRP), a well-established marker of the development of inflammation, tumor necrosis factor (TNF)-α, an inflammatory marker strongly associated with diabetes, and adiponectin, a cytokine produced by adipose tissue and associated with insulin sensitivity. these cytokines play a major role in the development of T2D.

Practical: Nutrition in autoimmune rheumatoid diseases and Sjogren syndrome

Objectives

Recognize How rheumatoid arthritis affect body composition.
Discuss the aims of nutritional treatment in patients with rheumatoid arthritis.
Recognize dietary modifications that are necessary for the management of xerostomia in patients with Sjogren syndrome.
List drugs used for the treatment of rheumatic diseases that can cause drug–nutrient interactions.

Is nutrition a predisposing factor for autoimmune rheumatoid diseases?

The long-term consumption of foods with pro-inflammatory properties, such as meat and the abstention from foods with anti-inflammatory properties, such as fish, fruits and vegetables, may promote the development of autoimmune diseases. However, scientific data have, so far, not managed to reveal such a relationship and attribute the predisposition to autoimmune diseases to other factors, namely genetic, hormonal, environmental or psychological.

How does rheumatoid arthritis affect body composition?

Rheumatoid cachexia was first described over a century ago. However, this 'bad condition' has been recognized as a common problem among patients with rheumatoid arthritis (RA) relatively recently. While cachexia generally connotes a state of advanced malnutrition and wasting, we now know that this term refers, more specifically, to a loss of body cell mass (BCM). TNF-alpha and IL-1-beta, which play the most important role in the pathogenesis of RA, affect total body protein significantly, by promoting protein breakdown and loss. Loss of BCM greater than 40% of baseline is associated with death. However, even with as little as 5% loss of BCM, there are demonstrable changes in morbidity, including loss of muscle strength, altered energy metabolism and increased susceptibility to infections.
Rheumatoid cachexia should be viewed as an important contributor to increased morbidity and premature mortality in RA and in two-thirds of patients is combined with obesity, and the state has been described as 'rheumatoid cachectic obesity'. Patients with RA usually present with increased fat deposition throughout the body, apart from in the legs; therefore, fat distribution becomes more central, and this is more prominent in patients receiving corticosteroids for long periods. In addition, low habitual physical activity has consistently been observed in RA and is an important consequence of, and contributor to, muscle wasting. Moreover, low physical activity

predisposes to fat gain and is believed to precipitate a negative reinforcing cycle of muscle loss, reduced physical function and fat gain in RA, which leads to 'cachectic obesity'.
Bone mineral density in several sites is also reduced in patients with RA, compared to healthy controls. This finding has been attributed to several factors, namely long-term corticosteroid use, low physical activity level, increased secretion of TNF-alpha and IL-1-beta which promote osteoclast differentiation and, additionally, the low intake of calcium and vitamin D.
Therefore, any nutritional assessment of patients with RA should always focus on the dietary intake of these nutrients.

What are the aims of nutritional treatment in patients with rheumatoid arthritis?

The main dietary goals for patients with RA are:
_ muscle mass preservation, through appropriate diet, especially in terms of protein intake, and exercise counselling
_ prevention of fat mass increase, especially at central body parts
_ emphasis on all the nutrients that optimise bone health (calcium, vitamin D, protein) and restriction of those with a detrimental effect (salt, alcohol, vitamin A)
_ diet adaptation according to medication (especially during corticosteroid use)
_ modification of diet texture according to chewing, swallowing and selffeeding ability.

Do elimination diets have a role in the treatment of rheumatoid arthritis?

Patients with RA often claim that their symptoms are alleviated by special diets or by simply eliminating certain foods, and it has been proposed that food-related antigens, predominantly from protein sources, may provoke hypersensitivity responses, which may increase symptoms of RA.
Elimination diets are usually preceded by a period of fasting, which may confound the reported improvement in symptoms. Fasting is known to suppress inflammation. The mechanism by which this operates is not completely understood, but may involve a reduction in the release of pro-inflammatory cytokines, reduced leukotriene formation and altered intestinal permeability, which may decrease the penetration of immunostimulants from the intestines.
Short-term fasting with or without a prolonged vegetarian diet may represent another interesting dietary approach in RA. The clinical effectiveness of this dietary approach, however, remains unknown, and possible anti-inflammatory mechanisms are far from understood. Fasting, elimination and elemental diets cannot be regarded as having an established place in treatment but may help individual patients.

objective parameters. Patients with RA should be discouraged from undertaking self-imposed elimination diets, which may compromise their nutritional status and should be advised to eat a well-balanced diet, according to their anthropometric and clinical characteristics and the medication they receive.

What should a dietitian bear in mind about the nutrition of a patient with systemic lupus erythematosus?

Patients with systemic lupus erythematosus (SLE) may present several and various symptoms, according to the affected organ or systems of the body. The most common nutrition-related complications of SLE are lupus nephritis, which is accompanied with urinary protein loss and electrolyte disturbances, hypertension and atherosclerosis. Therefore, each patient, according to their symptoms and problems, may follow a different diet. The dietitian should monitor patients' biochemical indices and symptoms and change the diet accordingly. Moreover, a patient's medication should be carefully monitored since it may influence the diet regime.

What dietary modifications are necessary for the management of xerostomia in patients with Sjogren syndrome?

Restriction of carbohydrates, especially of the simple ones like sugar and honey, as well as the food and commercially available products that contain them, can help patients with xerostomia. Moreover, hard and dehydrated food (e.g. rusks, Melba toast, crackers) or food at extreme temperatures should be avoided. Conversely, frequent cleaning of the teeth and the use of mouthwash are common practices that the patient could find helpful and should always bear in mind.

Which of the drugs used for the treatment of rheumatic diseases can cause drug–nutrient interactions or impose certain dietary modifications?

Corticosteroids are the cornerstone of treatment for several rheumatic diseases;
however, their long-term use may generate several adverse effects, some of which will affect the patient's diet. In order to avoid severe fluid retention when using corticosteroids, patients should follow a low-sodium diet. Moreover, corticosteroids may raise blood sugar or even cholesterol levels, and therefore patients should be advised to restrict the consumption of simple sugars or saturated fat, respectively. If a patient's blood glucose level rises significantly, they should restrict themselves to a diabetic diet while they are taking corticosteroids. In addition, since corticosteroids induce protein catabolism, patients should consume the necessary daily protein amount and should achieve an optimum dietary intake in terms of bone health, since long-term corticosteroid use comprises an important risk factor for osteoporosis.

Therefore, during corticosteroid treatment, it is vital to ensure the recommended calcium and vitamin D intake, either from foods or from supplements. Patients receiving large doses of corticosteroids usually report a weight gain, and this can be attributed entirely to the increase in appetite that the drug causes. Dietitians should inform their patients of this side effect and encourage them to follow a certain diet according to their energy needs and consumption and not according to their appetite. Methotrexate is a known antagonist of folic acid, and folic acid levels may be reduced in patients receiving this drug. The careful monitoring of folic acid levels is required, whereas a low weekly supplementation dose of folic acid (<5 mg) can prevent deficiency of this nutrient. Dietitians should bear in mind that patients receiving cyclosporin should completely avoid the consumption of grapefruit and its juice, since this fruit contains a factor that increases the drug's toxicity.

Can patients with rheumatic diseases benefit from alternative medicine supplements?

There are several over-the-counter dietary supplements, herbs or combinations of nutrients and herbs either from large reliable industries or from other, less safe, sources that claim several immunosuppressant or immunostimulatory properties. The most common supplements reported by patients with rheumatic diseases are fish oil or omega-3 polyunsaturated fatty acids, gamma-linolenic acid and antioxidants (e.g. C, E and selenium). The scientific evidence so far does not justify the systematic use of such supplements, since some of them may exhibit serious side effects or interactions with the recommended medication. The most evidence-based information exists for the use of fish oils as part of the treatment for inflammatory disorders such as RA; however, the recommended effective doses remain a controversial issue. Antioxidant supplementation, while seeming to have plausibility, lacks a comparable evidence base and the only dietary supplements that doctors routinely recommend to most patients with rheumatic diseases are calcium and vitamin D.

Chapter 7: Food Allergy

Objectives

Explain the role of nutrition in food allergy.
Discuss the main types of food allergy and how are they clinically presented.

Overview of the food allergy and related concepts

Food allergy refers to specific reactions that result from an abnormal immunological response to a food and which can be severe and life-threatening and triggered by minute amounts of the allergen.

Non-allergic food intolerance refers to reactions to food that can result from a number of causes, none of which is mediated by the immune system (e.g. pharmacological effects, enzyme deficiencies, irritant and toxic effects). Owing to their variable aetiology, their effects can be acute and severe, although rarely life-threatening, but they are usually chronic and diffuse. Unlike food allergy, relatively large amounts of a food are usually necessary for adverse effects to occur.

The main types of food allergy and how are they clinically presented

There are two major classes of food allergic reactions:

1-immunoglobulin E(IgE) mediated:

are generally present soon after ingestion and thus easy to investigate and diagnose. They can be more violent than non-IgE mediated reactions and can even lead to death through anaphylaxis in severe cases.

2- Non-IgE mediated reactions:

are often presented later and often an important cause of ill health. Food-allergic reactions are generally divided into those of early onset (within minutes to an hour after food ingestion), which also tend to be IgE mediated, and those of late onset (taking hours or days), which are in general non-IgE mediated. Early-onset manifestations often include wheezing, urticaria, angioedema, rashes, vomiting and anaphylaxis, whereas late-onset symptoms include diarrhoea, abdominal pain, allergic rhinitis, atopic eczema food-sensitive enteropathy or food-sensitive colitis, protein-losing enteropathy and constipation.

What foods are known to cause immunologically mediated reactions?

Common foods that can cause an allergic reaction include:

_ peanuts and tree nuts (e.g. hazelnut, Brazil nut, walnut)
_ milk (cow’s, goat’s, sheep’s)
_ soya
_ fish
_ shellfish

_ eggs
_ seeds (especially sesame and caraway)
_ fruits (especially apples, peaches, plums, cherries, bananas, citrus fruits)
_ herbs and spices (especially mustard, paprika and coriander).

However, the most common allergies, according to the frequency they occur, are:

_ in children: cow's milk, egg, soya, peanut, tree nuts, fish and crustaceans
_ in adults: peanut, tree nuts, crustaceans, fish and egg.

The processing of food may also affect its allergenicity. For example, the allergenicity of many fruits may be greatly reduced by cooking, and that of eggs, milk and some fish may be attenuated. It is also possible that the boiling, but not the roasting, of peanuts may lessen their allergenicity. On the other hand, the thermal processing of food may lead to the formation of allergens that are not present in raw foods. This may be attributed to changes in the shape of protein molecules and the revealing of previously hidden epitopes. Moreover, allergies to 'new' foods commonly emerge as these foods are introduced to a new population. In addition, novel food proteins from genetically modified organisms or from new manufacturing processes applied to existing foods could carry a risk of food allergy.

Practical: Diagnosis and management of food allergy

Objectives

Recognize the best tools for diagnosis of food allergy.
Discuss How can a dietitian contribute to the management of food allergies.

How can food allergy be diagnosed?

The diagnosis of IgE mediated food allergy is usually based on a patient's medical history and confirmed by the results of one or more specific investigations, including skin tests, blood tests [radioallergosorbent (RAST) tests, enzyme-linked immunosorbent assay (ELISA)], response to dietary restriction (single exclusion diet, multiple-food exclusion diet, elemental and protein hydrolysate formula diet) and sometimes by oral challenge tests. The confirmation of non-IgE mediated (delayed) food allergy is more difficult to achieve and is largely based on dietary restriction and oral challenge tests.

How can a dietitian contribute to the management of food allergies?

The management of allergic diseases is recognised as an area of specialization and should ideally be carried out by a clinical immunology and allergy team, which would include medical expertise from the fields of respiratory diseases, dermatology, gastroenterology and immunology and the specialist skills of dietitians and nurses. This team should identify and completely avoid the offending allergen or allergens. Specialist dietetic guidance is essential to ensure that

_ all potential sources of the allergen are avoided
_ the effects of the exclusion diet on the intake of other nutrients and overall dietary balance are minimised.

This is especially important for infants and children, and in the event that the excluded food is a major nutritional contributor (e.g. milk), because alternative sources of nutrients should be provided. Dietitians should also provide written guidance on the foods or types of foods that must be avoided, foods or types of foods that may need to be avoided, which is determined from ingredients lists of manufactured foods, and foods or types of foods that can be safely eaten. Dietary advice in order to prevent potential nutritional inadequacies is also crucial. Moreover, dietitians should teach their patients to:

_ carefully check all ingredient labels
_ learn other names of the food responsible for the allergy
_ exercise caution when eating out since restaurant staff are not always aware of specific menu ingredients or how food is prepared

_ be careful when eating food that is packaged in multi-packs with other foods: while one product may be considered safe, there is a risk of cross contamination because products may leak or become unwrapped.

Chapter 8: HIV/AIDS disease and nutrition

Objectives

List the main modes of transmission of HIV.
Relate the main factors precipitating weight loss and wasting to HIV disease.
Discuss main goals of nutritional care for HIV-positive patients.
Discuss the goals of the nutritional management of symptomatic patients with HIV infection.

Overview of HIV/AIDS disease and Related Concepts

What is HIV/AIDS disease and what are the main modes of transmission?

Acquired immunodeficiency syndrome (AIDS) is a disorder resulting from the infection with the human immunodeficiency virus (HIV), which leads to a profound immunosuppression and high susceptibility to life-threatening opportunistic infections and malignancies.

HIV infection can be transmitted by:

_ unprotected vaginal and anal sexual intercourse

_ receipt of infected blood and blood products (sharing injecting equipment between drug users, occupational accidents with needle stick injuries, infusion of contaminated blood products)

_ from the infected mother to the child during pregnancy, birth or breastfeeding (vertical transmission).

What are the main goals of nutritional care for HIV-positive patients?

Since malnutrition is one of the most common problems in patients with HIV/AIDS, nutritional care aims predominately at the efficient provision of nutrients and the maintenance of body weight and lean body mass within normal levels. Moreover, the delay of wasting syndrome and the early detection and treatment of the metabolic disturbances due to the disease or the antiretroviral therapy are of main concern. Finally, patients with HIV/AIDS should be provided with nutritional advice tailored to their specific needs, the possible side effects of their medications and their lifestyles.

Classification of HIV disease according to the Centers of Disease Control in the USA.

CD4 lymphocytes count	Symptoms A Asymptomatic disease, acute retroviral syndrome, generalised lymphadenopathy	B Symptoms of AIDS-related complex	C AIDS-defining conditions
>500/µL	A1	B1	C1
200–499/µL	A2	B2	C2
<200/µL	A3	B3	C3

All patients in categories C1, B2, C2, A3, B3 and C3 are characterised as patients with AIDS.

What are the main factors precipitating weight loss and wasting in HIV disease?

The most common nutritional problem in symptomatic HIV disease is weight loss or wasting. Although its severity and frequency appear to be reduced by the use of antiretroviral therapy, it remains a problem. Since malnutrition can reduce the quality of life and increase morbidity and mortality in these patients, the early detection and treatment of the underlying cause of insufficient nutrient intake is of vital importance.

Malnutrition in HIV patients can often be attributed to:

_ reduced food intake, due to psychological an neurological problems, sore mouth or side effects of their medication (i.e. anorexia, vomiting, nausea)

_ altered metabolic needs due to the progression of the disease

_ malabsorption, especially in patients with chronic diarrhoea caused by gastrointestinal pathogens, by medication or by the HIV disease itself.

What are the nutritional needs of an asymptomatic or weight stable patient with HIV infection?

The main goal for asymptomatic or weight stable HIV-positive patients is to optimise food intake and to ensure the adequate intake of nutrients. Unintentional weight loss should be avoided by the sufficient provision of energy and protein. Although the micronutrient requirements of HIV-positive patients are unknown, a multivitamin and mineral supplement, providing no more than 100% of dietary reference intake (DRI) or reference nutrient intake (RNI) could be prescribed in order to avoid deficiencies, especially when food intake is compromised.

What are the goals of the nutritional management of symptomatic patients with HIV infection?

The main goals of the nutritional management of symptomatic patients with HIV infection are:

_ the preservation or increase of lean body mass

_ adequate provision of macro- and micronutrients

_ achievement and/or maintenance of a body weight within the normal values for body mass index (BMI)

_ provision of symptomatic relief, according to the patient's needs.

How are the energy and protein needs in symptomatic HIV-positive patients calculated?

In being a part of the medical care team responsible for the management of HIV-positive patients, the clinical dietitian should have the ability to combine the available data from the nutritional assessment, the medical treatment and the patient's individual needs in order to provide tailor-made advice to the patient.

The energy needs of these patients vary greatly, according to the state of the disease, the presence of problems that could compromise absorption and the utilisation of nutrients. Ideally, energy needs should be calculated by indirect calorimetry. However, in the clinical setting, energy needs could be calculated, with limited accuracy though, by the use of equations such as the Harris-Benedict or Schofield equations, taking into account the relative stress factors according to the overall health status of the patient. Energy requirements are likely to increase by 10% to maintain body weight and physical activity in asymptomatic HIV-infected adults. During symptomatic HIV, and subsequently during AIDS, energy requirements increase by approximately 20–30% to maintain adult body weight. Protein needs in HIV-positive patients are considered relatively high. A dietary protein intake of 1.5–2 g/kg is considered adequate for the majority of patients. Likewise, providing 20% of a patient's daily energy intake as protein seems to cover the needs of HIV-infected patients, given the fact that their energy needs are high. These recommendations may differentiate in patients with kidney or hepatic disease. In these cases, the relevant recommendations for non-HIV patients should be followed.

Do vitamin and mineral needs differ in symptomatic HIV-positive patients?

The exact micronutrient needs of HIV-positive patients are not established yet. According to the available data, HIV-positive patients seem to have lower levels of vitamin A, B6, C and E in the bloodstream. Recently, possibly HIV-positive patients may need these vitamins more than the healthy population does.

Practical: Nutrition-related side effects of highly active antiretroviral therapy

Objectives

Recognize the main nutrition-related side effects of highly active antiretroviral therapy.
Discuss the role of nutrition in the management of HAART-induced dyslipidemia.

What is highly active antiretroviral therapy?

'Highly active antiretroviral therapy' (HAART) is a term used to describe the use of a combination of antiretroviral drugs for the treatment of HIV infection. The main antiretroviral drugs that are used for HIV-positive subjects are categorised according to the phase of the retrovirus life cycle that the drugs inhibit, as follows:

_ nucleoside and non-nucleoside reverse transcriptase inhibitors (NRTIs and nNRTIs respectively) inhibit reverse transcription of the virus.
_ protease inhibitors (PIs) inhibit the activity of protease (an enzyme used by HIV to cleave nascent proteins for the final assembly of new virons).
_ integrase inhibitors inhibit integrase, the enzyme that is responsible for the integration of viral DNA into the DNA of the host cell.
_ fusion inhibitors interfere with the binding, fusion and entry of the HIV virus to the host cell.
_ maturation inhibitors inhibit the last step of virus production, by blocking the formation of the capsid protein of the virus.

What are the main nutrition-related side effects of highly active antiretroviral therapy?

The use of HAART has transformed HIV infection from an acute illness to a manageable chronic condition. However, the significant decrease in mortality of HIV patients and the increase in their life expectancy have been accompanied by several clinical and metabolic complications.

Nutrition-related side effects have been shown to correlate negatively with quality-of-life measures in people infected with HIV. The most common gastrointestinal adverse effects include nausea, vomiting, dyspepsia, anorexia, abdominal pain, chewing/swallowing difficulties and taste alterations. Usually, these effects lose their intensity after the first month of treatment. If they persist, they may compromise dietary intake, and a change in the drug regimen should be considered. Providing specific strategies to support patients through these challenges is an important part of nutritional therapy.

The main metabolic complication is described as 'HAART-induced lipodystrophy syndrome', which includes:

_ lipoatrophy (face, extremities and buttocks)
_ fat accumulation (abdomen, dorsocervical or supraclavicular fat pad – buffalo hump)
_ dyslipidaemia [hypertriglyceridaemia, hypercholesterolaemia and high-density lipoprotein (HDL) cholesterol]
_ insulin resistance and glucose intolerance
_ lactic acidosis.

What is the role of nutrition in the management of HAART-induced dyslipidaemia?

The current recommendations for the management of dyslipidaemia in HIVinfected patients receiving HAART do not differ from the recommendations for the non-HIV-infected population.

Adult Treatment recommendations, which include dietary restriction of total fat to 25–35% of total energy intake, saturated fat to less than 7% of total calories, dietary cholesterol to less than 200mg per day, use of plant sterols (2 g/d) and attainment of a prudent diet increased with a high intake of dietary fibre (10–25 g/d). Recent data also support the use of a Mediterranean diet pattern (low in saturated fat, without a severe restriction of monounsaturated or omega-3 polyunsaturated fat, replacing some of the complex carbohydrates with monounsaturated fatty acids). Increased physical activity or exercise, especially weight-bearing exercise in combination with aerobic activities, could benefit patients, by reserving muscle mass and improving cardiovascular health. Lifestyle changes recommended for the treatment of isolated hypertriglyceridaemia, in addition to reduced fat intake and exercise, include reduction in alcohol and refined-sugars intake.

What is AIDS wasting syndrome and how can it be managed?

'Wasting syndrome' is a term used to describe unintentional weight loss (>10% of the usual body weight) in combination with diarrhoea, fatigue and/or fever for a period of more than 30 days. The weight loss concerns mainly the loss of lean body mass and, secondarily, fat mass. Wasting can contribute to the deterioration of a patient's immune system and is connected with high rates of mortality and usually signals that the HIV-infection is progressing to AIDS.

Detailed dietary assessment should be performed regularly in HIV-infected patients to ensure the early detection of wasting syndrome. When and if it is identified, certain actions should take place in order to maximise dietary intake of the patient and halt their weight loss. Dietary advice should focus on energy- and nutrient-dense foods, while meal planning should be the first priority for these patients. Food enrichment and food fortification could also improve the nutrient intake. If the patient's dietary intake remains compromised, oral supplements could be used.

If the patient fails to attain a sufficient dietary intake, the use of artificial nutrition should be considered. Enteral and parenteral nutrition could be used in order to preserve the patient's nutritional status.

Does omega-3 fatty acid intake benefit patients with HIV infection?

Omega-3 fatty acids have been shown to be effective for lowering triglycerides in patients with HIV-associated dyslipidaemia. Omega-3 fatty acids may also have secondary benefits in decreasing bone resorption and decreasing markers of systemic inflammation.

What are the main strategies for reducing the danger of food- and water-borne illnesses?

HIV-infected patients, owing to their compromised immune system, are usually more vulnerable to food-borne illness than the non-infected population. Special care should be taken for the education of HIV-infected patients in the safe handling of food in order to minimize the danger of a food-borne illness. Strict hygiene measures should be taken during food preparation, and foods easily spoilt or of a high microbial load should be avoided (raw or semi-raw meat and fish, raw eggs, non-pasteurised milk and milk products, soft cheese and cheese with molds, i.e. Roquefort or blue cheese). Moreover, attention should be paid to the expiry dates of food products and to the careful storing

of food. In the developed world, tap water is usually safe. If water comes from wells or rivers, it should be boiled before it is consumed. Ice cubes and cold drinks should also be prepared from safe water in order to avoid water-borne illnesses.

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Chapter 9: Microorganisms in human welfare: probiotics

Objectives

Define probiotic and single cell protein.
Recognize the selection criteria for probiotic strains.
List the commonest Probiotic Microorganisms.
Outline different Mechanisms of Action of Probiotics.
List The beneficial effect of probiotics.

Overview of probiotics and related concepts

Probiotics are microorganisms that are claimed to provide health benefits when consumed.

Single-cell protein (**SCP**) refers to edible unicellular microorganisms. The biomass or protein extract from pure or mixed cultures of algae, yeasts, fungi or bacteria may be used as an ingredient or a substitute for protein-rich foods, and is suitable for human consumption or as animal feeds.

Selection Criteria and Requirements for Probiotic Strains

According to the suggestions of the WHO (world health organization) and EFSA (the European Food Safety Authority), in their selection process, probiotic strains must meet both safety and functionality criteria

The safety of a strain is defined by its origin, the absence of association with pathogenic cultures, and the antibiotic resistance profile. Functional aspects define their survival in the gastrointestinal tract and their immunomodulatory effect. Probiotic strains have to meet the requirements associated with the technology of their production, which means they have to be able to survive and maintain their properties throughout the storage and distribution processes.

Probiotics should also have documented pro-health effects consistent with the characteristics of the strain present in a marketed product.

Selection criteria of probiotic strains

Criterion	Required Properties
Safety	• Human or animal origin. • Isolated from the gastrointestinal tract of healthy individuals. • History of safe use. • Precise diagnostic identification (phenotype and genotype traits). • Absence of data regarding an association with infective disease. • Absence of the ability to cleave bile acid salts. • No adverse effects. • Absence of genes responsible for antibiotic resistance localised in non-stable elements.
Functionality	• Competitiveness with respect to the microbiota inhabiting the intestinal ecosystem. • Ability to survive and maintain the metabolic activity, and to grow in the target site. • Resistance to bile salts and enzymes. • Resistance to low pH in the stomach. • Competitiveness with respect to microbial species inhabiting the intestinal ecosystem (including closely related species). • Antagonistic activity towards pathogens (e.g., *H. pylori*, *Salmonella* sp., *Listeria monocytogenes*, *Clostridium difficile*). • Resistance to bacteriocins and acids produced by the endogenic intestinal microbiota. • Adherence and ability to colonise some particular sites within the host organism, and an appropriate survival rate in the gastrointestinal system.
Technological usability	• Easy production of high biomass amounts and high productivity of cultures. • Viability and stability of the desired properties of probiotic bacteria during the fixing process (freezing, freeze-drying), preparation, and distribution of probiotic products. • High storage survival rate in finished products (in aerobic and micro-aerophilic conditions). • Guarantee of desired sensory properties of finished products (in the case of the food industry). • Genetic stability. • Resistance to bacteriophages.

Probiotic Microorganisms

Probiotic products may contain one or more selected microbial strains. Human probiotic microorganisms belong mostly to the following geni: Lactobacillus, Bifidobacterium, and Lactococus, Streptococcus, Enterococcus. Moreover, strains of Gram-positive bacteria belonging to the genus Bacillus and some yeast strains belonging to the genus Saccharomyces are commonly used in probiotic products .

Probiotics are subject to regulations contained in the general food law, according to which they should be safe for human and animal health.

probiotic microorganisms can be contained in pharmaceutical products and as food additives.

Probiotic microorganisms used in human nutrition

Type *Lactobacillus*	Type *Bifidobacterium*	Other Lactic Acid Bacteria	Other Microorganisms
L. acidophilus (a),* *L. amylovorus* (b),* *L. casei* (a),(b),* *L. gasseri* (a),* *L. helveticus* (a),* *L. johnsonii* (b),* *L. pentosus* (b),* *L. plantarum* (b),* *L. reuteri* (a),* *L. rhamnosus* (a),(b),*	*B. adolescentis* (a) *B. animalis* (a),* *B. bifidum* (a) *B. breve* (b) *B. infantis* (a) *B. longum* (a),*	*Enterococcus faecium* (a) *Lactococcus lactis* (b),* *Streptococcus thermophilus* (a),*	*Bacillus clausii* (a),* *Escherichia coli Nissle 1917* (a) *Saccharomyces cerevisiae (boulardi)* (a),*

(a) Mostly as pharmaceutical products; (b) mostly as food additives; * QPS (Qualified Presumption of Safety) microorganisms.

Mechanism of Action of Probiotics

-Probiotics have numerous advantageous functions in human organisms. Their main advantage is the effect on the development of the microbiota inhabiting the organism in the way ensuring proper balance between pathogens and the bacteria that are necessary for a normal function of the organism.

-Live microorganisms meeting the applicable criteria are used in the production of functional food and in the preservation of food products. Their positive effect is used for the restoration of natural microbiota after antibiotic therapy. Another function is counteracting the activity of pathogenic intestinal microbiota, introduced from contaminated food and environment.Therefore, probiotics may effectively inhibit the development of pathogenic bacteria, such as Clostridium perfringens, Campylobacter jejuni, Salmonella Enteritidis, Escherichia coli, various species of Shigella, Staphylococcus, and Yersinia, thus preventing food poisoning.

-A positive effect of probiotics on digestion processes, treatment of food allergies and dental caries. Probiotic microorganisms such as Lactobacillus plantarum, Lactobacillus reuteri, Bifidobacterium adolescentis, and Bifidobacterium pseudocatenulatum are natural producers of B group vitamins (B1, B2, B3, B6, B8, B9, B12). They also increase the efficiency of the immunological system, enhance the absorption of vitamins and mineral compounds, and stimulate the generation of organic acids and amino acids. Probiotic microorganisms may also be able to produce enzymes, such as esterase, lipase, and co-enzymes.Some products of probiotics' metabolism may also show antibiotic (acidophiline, bacitracin, lactacin), anti-cancerogenic, and immunosuppressive properties.

The beneficial effect of probiotics, involving four mechanisms:

(1) Antagonism through the production of antimicrobial substances.
(2) Competition with pathogens for adhesion to the epithelium and for nutrients.
(3) Immunomodulation of the host.
(4) Inhibition of bacterial toxin production.

The first two mechanisms are directly associated with their effect on other microorganisms. Those mechanisms are important in prophylaxis and treatment of infections, and in the maintenance of balance of the host's intestinal microbiota. The ability of probiotic strains to co-aggregate, as one of their mechanisms of action, may lead to the formation of a protective barrier preventing pathogenic bacteria from the colonization of the epithelium. Probiotic bacteria may be able to adhere to epithelial cells, thus blocking pathogens. That mechanism exerts an important effect on the host's health condition. Also a possible role of probiotics in the elimination of cancer cells.

The immunomodulatory effect of the intestinal microbiota, including probiotic bacteria, is based
on three, seemingly contradictory phenomena:

(1) Induction and maintenance of the state of immunological tolerance to environmental antigens (nutritional and inhalatory);
(2) Induction and control of immunological reactions against pathogens of bacterial and viral origin;
(3) Inhibition of auto-aggressive and allergic reactions.

Practical: Importance of microbes in food biotechnology

Objectives

Recognize use of Lactobacilli in the food industry.
Recognize use of Saccharomyces sps in the food industry.
Outline the beneficial effect of probiotics for humans.

Microorganisms, particularly the bacteria and fungi, have served humans since hundreds of years for the purpose of food, drugs, and other high-value chemical products. The use of microbes for fermentation is known since Neolithic age. Microbes not only give a good taste, texture and smell to the foods, but also produce certain inhibitory compounds that help in stopping food spoilage thus increasing the storage and safety of food.

The use of Lactobacilli in the food industry:

Lactobacilli are important in the production of foods that require lactic acid fermentation, notably dairy products [yogurt and cheese], fermented vegetables [olives, pickles, and sauerkraut], fermented meats [salami], and sourdough bread.

The genera important members of this group are *Lactobacillus*, *Leuconostoc*, *Pediococcus* and *Streptococcus*. These organisms are heterotrophic and generally have complex nutritional requirements due to lacking of many biosynthetic capabilities.

The use of Saccharomyces sps in the food industry:

Saccharomyces sps. have found historic use in baking and brewing industry. Particularly *S. cerevisiae* plays an important role in revenue generation from brewing industry. Other fungi such as *Aspergillus* sps. and *Penicillium* sps. have also found use in food industry particularly due to the enzymes obtained from them. Fungi have an important role in probiotics preparation as well. The best example of drug production by microbes lies in antibiotics production.

Another major class of pharmaceutical products obtained from microbes is of recombinant proteins where eukaryotic production system such as yeast is preferred over prokaryotic system due to its ability to carry out post translational modifications in the expressed proteins. The approved protein products produced exclusively in *Saccharomyces cerevisiae* are hormones [insulin, insulin analogues, non-glycosylated human growth hormone somatotropin, glucagon], vaccines (hepatitis B virus surface antigen)

Probiotics for Humans:

In the face of widespread diseases and ageing societies, the use of knowledge on microbiocenosis of the Probiotics for Humans In the face of widespread diseases and ageing societies, the use of knowledge on microbiocenosis of the gastrointestinal tract and on the beneficial effect of probiotic bacteria is becoming increasingly important. The consumption of pre-processed food (fast food), often containing excessive amounts of fat and insufficient amounts of vegetables, is another factor of harmful modification of human intestinal microbiota. There is currently no doubt about the fact that the system of intestinal microorganisms and its desirable modification with probiotic formulas and products may protect people against enteral problems, and influence the overall improvement of health.

-Probiotics may be helpful in the treatment of inflammatory enteral conditions, including ulcerative colitis, Crohn’s disease, and non-specific ileitis.

-Consumption of probiotics-containing dairy products results in the reduction of blood cholesterol, which may helpful in the prevention of obesity, diabetes, cardiovascular diseases, and cerebral stroke. The reduction of cholesterol level achieved due to probiotics is less pronounced compared to the effect of pharmaceutical agents, but leads to a significant minimisation of side effects.

-The application of probiotics in the treatment of diarrhoea. The application of Saccharomyces boulardii yeast to patients with acute, watery diarrhoea resulted in the cure and reduced frequency of that type of complaints.

-Lactobacillus rhamnosus in the form of a probiotic resulted in a reduced number of respiratory tract infections.

Chapter 10: Prebiotic

Objectives

Define prebiotics and Synbiotics.
Recognize the selection criteria for prebiotic.
List the commonest Prebiotic.
Outline different Mechanisms of Action of Prebiotics.
List The beneficial effect of prebiotics for humans.

Overview of prebiotics and related concepts

Prebiotics are food ingredients that induce the growth or activity of beneficial microorganisms (bacteria and fungi). The most common example is in the gastrointestinal tract, where prebiotics can alter the composition of organisms in the gut microbiome.

Dietary prebiotics are typically non-digestible fiber compounds that pass undigested through the upper part of the gastrointestinal tract and stimulate the growth or activity of advantageous bacteria that colonize the large bowel by acting as substrate for them. As a functional food component, prebiotics, like probiotics, are conceptually intermediate between foods and drugs. Depending on the jurisdiction, they typically receive an intermediate level of regulatory scrutiny, in particular of the health claims made concerning them.

Synbiotics are food ingredients or dietary supplements combining probiotics and prebiotics in a form of synergism, hence synbiotics. The synbiotic concept was first introduced as "mixtures of probiotics and prebiotics that beneficially affect the host by improving the survival and implantation of live microbial dietary supplements in the gastrointestinal tract, by selectively stimulating the growth and/or by activating the metabolism of one or a limited number of health-promoting bacteria, thus improving host welfare"

The commonest prebiotics:

Fruit, vegetables, cereals, and other edible plants are sources of carbohydrates constituting potential prebiotics. The following may be mentioned as such potential sources: tomatoes, artichokes, bananas, asparagus, berries, garlic, onions, chicory, green vegetables, legumes, as well as oats, linseed, barley, and wheat. Some artificially produced prebiotics are, among others: lactulose, galactooligosaccharides, fructooligosaccharides, maltooligosaccharides, cyclodextrins, and lactosaccharose. Lactulose constitutes a significant part of produced oligosaccharides (as much as 40%). Fructans, such as inulin and oligofructose, are believed to be the most used and effective in relation to many species of probiotics.

An endogenous source of prebiotics in humans is human breast milk, which contains oligosaccharides structurally similar to galactooligosaccharides (GOS), referred to as human milk oligosaccharides (HMOs). These HMOs were found to increase the Bifidobacteria bacterial population in breastfed infants, and to strengthen the infant immune system. Furthermore, HMOs play a role in the establishment of a healthy intestinal microbiota composition of newborns

Prebiotic Selection Criteria

There are five basic criteria for the classification of food components such as prebiotics:

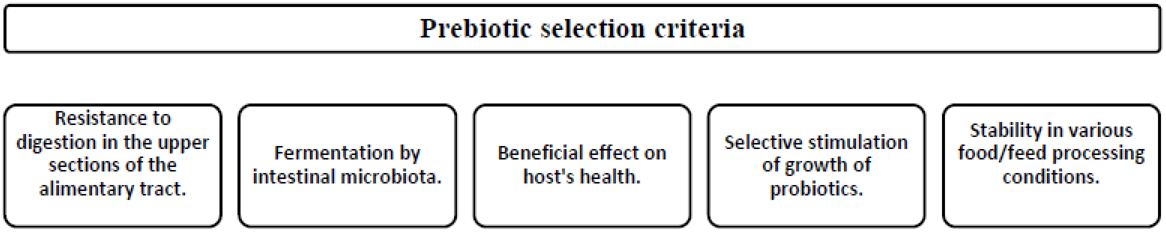
1-prebiotics are not digested (or just partially digested) in the upper segments of the alimentary tract. As a consequence, they reach the colon, where they are selectively fermented by potentially beneficial bacteria.

2-The fermentation may lead to the increased production or a change in the relative abundance of different short-chain fatty acids, increased stool mass, a moderate reduction of colonic pH, reduction of nitrous end products and fecal enzymes.

3- an improvement of the immunological system, which is beneficial for the host.

4-Slective stimulation of growth and/or activity of the intestinal bacteria potentially associated with health protection and wellbeing is considered another criterion.

5- a prebiotic must be able to withstand food processing conditions and remained unchanged, non-degraded, or chemically unaltered and available for bacterial metabolism in the intestine.



Prebiotics may be used as an alternative to probiotics or as an additional support for them. Long-term stability during the shelf-life of food, drinks, and feed, resistance to processing, and physical and chemical properties that exhibit a positive effect on the flavour and consistence of products may promote prebiotics as a competition to probiotics. Additionally, resistance to acids, proteases, and bile salts present in the gastrointestinal tract may be considered as other favourable properties of prebiotics. Prebiotic substances selectively stimulate microorganisms present in the host's intestinal ecosystem, thus eliminating the need for competition with bacteria. Moreover, prebiotics cause a reduction of intestinal pH and maintain the osmotic retention of water in the bowel. However, it should be considered that an overdose of prebiotics may lead to flatulence and diarrhoea—these effects are absent in the case of excessive consumption of probiotics. Prebiotics may be consumed on a long-term basis and for prophylactic purposes. Moreover, when used at correct doses, they do not stimulate any adverse effects, such as diarrhoea, or hepatic injuries caused by antibiotics. Prebiotic substances are not allergenic and do not proliferate the abundance of antibiotic-resistance genes. Of course, the effect of the elimination of selected pathogens achieved by the use of prebiotics may be inferior to antibiotics, so these properties make them a natural substitute for antibiotics.

Prebiotic Substances

The Most identified prebiotics are carbohydrates of various molecular structures, naturally occurring in human and animal diets. The physiological properties of potential prebiotics determine their beneficial effect on the host's health. Prebiotics may be classified according to those properties
as

_ not digested (or only partially digested);
_ not absorbed in the small intestine;
_ poorly fermented by bacteria in the oral cavity;
_ well fermented by seemingly beneficial intestinal bacteria;
_ poorly fermented by potential pathogens in the bowel.

Mechanism of Action of Prebiotics

Prebiotics are present in natural products, but they may also be added to food. The purpose of these additions is to improve their nutritional and health value. Some examples are: inulin, fructo oligosaccharides, lactulose, and derivatives of galactose and *b*-glucans. Those substances may serve as a medium for probiotics. They stimulate their growth, and contain no microorganisms. Prebiotics are not digested by host enzymes and reach the colon in a practically unaltered form, where they are fermented by saccharolytic bacteria (e.g., Bifidobacterium genus). The consumption of prebiotics largely affects the composition of the intestinal microbiota and its metabolic activity. This is due to the modulation of lipid metabolism, enhanced absorbability of calcium, effect on the immunological system, and modification of the bowel function. It is highly probable that providing an energy source that only specific species in the microbiota can utilize has a greater impact on microbiota composition and metabolism than these other factors.

The main aim of prebiotics is to stimulate the growth and activity of beneficial bacteria in the gastrointestinal tract, which confers a health benefit on the host.

Prebiotics for Humans

The presence of prebiotics in the diet may lead to numerous health benefits.

-the colorectal carcinoma occurs less commonly in people who often eat vegetables and fruit. This effect is attributed mostly to inulin and oligofructose.

-The reduction of the blood LDL (low-density lipoprotein) level, stimulation of the immunological system, increased absorbability of calcium, maintenance of correct intestinal pH value, low caloric value, and alleviation of symptoms of peptic ulcers and vaginal mycosis.

-The prevention of carcinogenesis, as well as the support of lactose intolerance or dental caries treatment.

Practical: Dairy products and their health benefits

Objectives

Recognize the health benefits of natural yoghurt.
Recognize the health benefits of cheese.
Outline different adverse effects of yoghurt.

Yogurt is a popular dairy product that's made by the bacterial fermentation of milk.

The bacteria used to make yogurt are called "yogurt cultures," which ferment lactose, the natural sugar found in milk.

This process produces lactic acid, a substance that causes milk proteins to curdle, giving yogurt its unique flavor and texture.

Yogurt can be made from all types of milk. Varieties made from skim milk are considered fat-free, whereas those made from whole milk are considered full-fat.

Plain yogurt without added colorants is a white, thick liquid with a tangy flavor.

Unfortunately, most commercial brands contain added ingredients, such as sugar and artificial flavors. These yogurts are not good for your health.

On the other hand, plain, unsweetened yogurt offers many health benefits.

Health benefits of natural yoghurt:

1-Yoghurt provides almost every nutrient that the body needs. It is especially high in calcium, B vitamins and trace minerals:

- It contains a lot of calcium, a mineral necessary for healthy teeth and bones. Just one cup provides 49% of the daily calcium needs.
- It is also high in B vitamins, particularly vitamin B12 and riboflavin, both of which may protect against heart disease and certain neural tube birth defects.
- One cup also provides 38% of the daily need for phosphorus, 12% for magnesium and 18% for potassium. These minerals are essential for several biological processes, such as regulating blood pressure, metabolism and bone health.

- One nutrient that yogurt does not contain naturally is vitamin D, but it is commonly fortified with it. Vitamin D promotes bone and immune system health and may reduce the risk of some diseases, including heart disease and depression.

2- Yoghurt is very high in protein. Protein is helpful for appetite and weight control:

- Yogurt provides an impressive amount of protein, with about 12 grams per 200 grams.
- Protein has been shown to support metabolism by increasing your energy expenditure, or the number of calories that you burn throughout the day.
- Getting enough protein is also important for appetite regulation, as it increases the production of hormones that signal fullness. It may automatically reduce the number of calories consumed overall, which is beneficial for weight control.
- Yoghurt has been shown to influence appetite control and delay feelings of hunger more than regular yoghurt with less protein.

3- Some types of yoghurt contain probiotics, which may boost digestive health by reducing the symptoms of common gastrointestinal disorders, such as bloating, diarrhea and constipation.

- Some types of yogurt contain live bacteria, or probiotics, that were either a part of the starter culture or added after pasteurization. These may benefit digestive health when consumed.
- Unfortunately, many yogurts have been pasteurized, which is a heat treatment that kills the beneficial bacteria they contain. So yoghurt that contains effective probiotics, should contains live, active cultures, which should be listed on the label.
- Some types of probiotics found in yogurt, such as *Bifidobacteria* and *Lactobacillus*, have been shown to lessen the uncomfortable symptoms of irritable bowel syndrome (IBS), which is a common disorder that affects the colon.
- Furthermore, probiotics may protect against antibiotic-associated diarrhea, as well as constipation.

4- Yogurt provides probiotics, vitamins and minerals, all of which may boost immune health and prevent certain illnesses:

- Consuming yogurt especially if it contains probiotics on a regular basis may strengthen the immune system and reduce the likelihood of contracting an illness.
- Probiotics have been shown to reduce inflammation, which is linked to several health conditions ranging from viral infections to gut disorders.
- Moreover, the immune-enhancing properties of yogurt are partly due to its magnesium, selenium and zinc, which are trace minerals known for the role they play in immune system health.
- Vitamin D-fortified yogurts may boost immune health even further. Vitamin D has been studied for its potential to prevent illnesses such as the common cold and flu.

5- Yoghurt is rich in vitamins and minerals that play a key role in bone health. Consuming it regularly may reduce the risk of osteoporosis.

- Yoghurt contains some key nutrients for maintaining bone health, including calcium, protein, potassium, phosphorus and, sometimes, vitamin D.
- All of these vitamins and minerals are especially helpful for preventing osteoporosis, a condition characterized by weakening of the bones. Which is common in the elderly.

6- Yoghurt appears to benefit heart health by increasing "good" high density lipoproteins (HDL) cholesterol and reducing blood pressure:

- Yogurt's fat content is one of the reasons why its healthiness is often controversial. It contains mostly saturated fat, with a small amount of monounsaturated fatty acids.
- There is no clear evidence that the fat in yogurt is harmful to health. In fact, it may benefit heart health.
- The intake of saturated fat from whole-milk products increases "good" HDL cholesterol, which may protect heart health. Other studies have found yogurt intake to reduce the overall incidence of heart disease.
- Furthermore, yogurt have been shown to help reduce high blood pressure, which is a major risk factor for heart disease. The effects seem to be most prominent in those already diagnosed with high blood pressure.

7-Yoghurt Promote Weight Management

- It is high in protein, which works along with calcium to increase levels of appetite-reducing hormones.
- The intake of full-fat dairy products, including yogurt, may reduce the incidence of obesity.

Adverse effects of yoghurt:

Yogurt may have adverse effects for those with lactose intolerance or milk allergies. Many types also contain high amounts of added sugar, which may contribute to certain health conditions.

Cheese

Cheese is rich in calcium, potassium, phosphorus, iron, and protein. Certain varieties are also rich in vitamins and good bacteria. We will further elaborate how cheese consumption can benefit the human body in 13 ways

1. Source of Protein

Protein is necessary for the body to maintain many functions, from a healthy metabolism to muscle regeneration. The protein present in cheese is easily digestible and is especially useful in the metabolic process. Protein is also an excellent source of energy and is one of the most crucial requirements of the human body. Cheese is an excellent source of protein and one way to utilize that is by adding it to your post-workout snack or on top of a salad. Cheddar and Swiss cheese compliments whole grain crackers quite well and feta can make for an excellent topping on a Greek salad.

2. Source of Carbohydrates

Carbohydrates are fuel for the body. The human body runs on Carbohydrates and requires them for the generation of energy. Cheese contains milk carbohydrates that break down in the form of glucose and sugar.

3. Fats

Cheese can be a source of good fat. The omega 3 and 6, along with the amino acids present in cheese can improve nerve and brain function. The body requires cholesterol in the form of good fat to carry out its daily procedures to prevent diseases.

4. Healthy Bones

Depending on our level of activity, our bones are prone to losing their health quite quickly. cheese can be an excellent addition to diet. This is because certain varieties are not only rich in calcium but also vitamin B and B complex. Vitamin B helps distribute the calcium throughout the body. Cheese is a good source of calcium and minerals for growing children.

5. A Healthy Heart

Certain varieties of it are good for the functions of the heart. cheese contains potassium, phosphorus, and magnesium which are a winning combination for your cardiovascular system. However, moderation is key to using cheese as a beneficial food.

6. Possible Cancer Prevention

It can contribute to reducing the risk of contracting cancer. This because it is rich in linoleic acid and sphingolipids, which are chemicals known for their antioxidant properties.

7. Management and Prevention of Osteoporosis

This illness triggers symptoms such as bone density loss and joint pains. In women, this is known to occur shortly after they experience menopause. But, this can be prevented by keeping a steady intake of cheese and other dairy products.

8. Healthy Teeth

Calcium and phosphorus make a winning combination for dental health. The calcium and other minerals present in cheese help maintain the strength and integrity of teeth.

9. Stress Reduction

Cheese has been observed to reduce panic and anxiety attacks. Where this can be attributed to the varieties rich n magnesium.

10. Probiotic Nature

We all know the benefits of yogurt as a probiotic, but it is not the only dairy byproduct that contains good bacteria. The good bacteria present in cheese help maintain gut health.

11. Brain Function

For healthy brain activity, Being rich in omega 3 and six fatty acids, cheese is a delicious way to keep the mind working.

12. Protein For Vegetarians

Vegetarians do not prefer eating meat or animal products, may find it difficult to keep up with their protein intake. Fortunately, cheese presents a great alternative source.

13. Immunity

Varieties that are rich in zinc are very beneficial for the body. They help keep the immune system strong. The good bacteria in cheese also help strengthen the digestive system from within which results in overall health and an increased ability to fight disease and infection

Chapter 11: Food and water borne diseases

Objectives

Recognize different mechanisms of food borne diseases.
List the commonest causative agents of foodborne illness.
Recognize clinical manifestations of food and water borne diseases.
Outline different preventive measures of foodborne illness.

Overview of Food borne diseases and related concepts

80-90% of Foodborne illnesses from Bacteria come from just 4 Bacteria
•Campylobacter
•Salmonella
•Clostridium perfringens
•Staphylococcus aureus

Illness Mechanisms:

•Infection
–Microorganisms are ingested and then cause illness
•Intoxication
–Toxins are produced by the pathogen, usually in the food. When food is consumed, illness occurs.
–Even if microorganisms are killed, toxin can still remain the food.

Bacterial Pathogens of Concern

-E. coli O157:H7

•Hemorrhagic colitis
•Cause: infection
•Incubation: 2-4 days
•Symptoms: diarrhea (blood),hemolytic uremic syndrome(HUS)
•Contaminant: milk, meat, fruits, vegetables, water

- Salmonella

•>2000 strains, 10 = foodborne illness
•Cause: infection
•Incubation: 6-48 hours
•Symptoms: nausea, fever, diarrhea, arthritis
•Contaminant: milk, meat, eggs

- Listeria monocytogenes

•Cause: infection
•Incubation: 2 days -3 weeks
•Symptoms: vomiting, diarrhea, meningitis, septicemia, miscarriage

•Contaminant: vegetables, milk, cheese, meat, seafood

-Campylobacter jejuni

•Cause: infection

•Incubation: 2 -5 days

•Symptoms: nausea, fever, diarrhea (blood)

•Contaminant: milk, meat, water.

- Staphylococcus aureus

•Cause: intoxication– (1 mg toxin = 100,000 cfu/g)

•Incubation: 1-6 hours

•Symptoms: nausea, fever, diarrhea

•Contaminant: milk, meat, egg

-Clostridium botulinum (Botulism)

•Cause: intoxication (spores -neurotoxin)

•Incubation: 18 -36 hours

•Symptoms: weakness, vertigo, difficulty in speaking, swallowing, breathing

•Contaminant: pH >4.6, low oxygen foods

Foodborne pathogens

Enteric viruses

Food and waterborne viruses contribute to a substantial number of illnesses throughout the world. Among those most commonly known are hepatitis A virus, rotavirus, astrovirus, enteric adenovirus, hepatitis E virus, and the human caliciviruses consisting of the noroviruses and the Sapporo viruses. This diverse group is transmitted by the fecal-oral route, often by ingestion of contaminated water and food.

Mycotoxins

Molds produce mycotoxins, which are secondary metabolites that can cause acute or chronic diseases in humans when ingested from contaminated foods. Potential diseases include cancers and tumors in different organs (heart, liver, kidney, nerves), gastrointestinal disturbances, alteration of the immune system, and reproductive problems Mycotoxins

occur mainly in cereal grains (barley, maize, rye, wheat), coffee, dairy products, fruits, nuts and spices. Control of mycotoxins in foods has focused on minimizing mycotoxin production in the field, during storage or destruction once produced.

Aflatoxins

are still recognized as the most important mycotoxins. The expression of aflatoxin-related diseases is influenced by factors such as age, nutrition, sex, species and the possibility of concurrent exposure to other toxins. The main target organ in mammals is the liver, so aflatoxicosis is primarily a hepatic disease. Conditions increasing the likelihood of aflatoxicosis in humans include limited availability of food, environmental conditions that favor mold growth on foodstuffs, and lack of regulatory systems for aflatoxin monitoring and control.

Yersinia enterocolitica

includes pathogens and environmental strains that are ubiquitous in terrestrial and fresh water ecosystems. Y. enterocolitica is a foodborne pathogen. Pork is often implicated as the source of infection. The pig is the only animal consumed by man that regularly harbors pathogenic Y.enterocolitica. An important property of the bacterium is its ability to multiply at temperatures near 0°C, and therefore in many chilled foods.

Vibrio

species are prevalent in estuarine and marine environments, and seven species can cause foodborne infections associated with seafood. Vibrio cholerae O1 and O139 serotypes produce cholera toxin and are agents of cholera. However, fecal-oral route infections in the terrestrial environment are responsible for epidemic cholera.

Staphylococcus aureus

is a common cause of bacterial foodborne disease worldwide. Symptoms include vomiting and diarrhea that occur shortly after ingestion of S. aureus toxin-contaminated food. The symptoms arise from ingestion of preformed enterotoxin, which accounts for the short incubation time.

Campylobacter

is one of the major causes of bacterial gastroenteritis worldwide. Campylobacter infection is primarily a foodborne illness, usually without complications; however, serious sequelae, such as Guillain-Barre Syndrome, occur in a small subset of infected patients.

Listeria monocytogenes

is Gram-positive foodborne bacterial pathogen and the causative agent of human listeriosis. Listeria infections are acquired primarily through the consumption of contaminated foods, including soft cheese, raw milk, deli* salads, and ready-to-eat foods such as luncheon meats and frankfurters. Although L. monocytogenes infection is usually limited to individuals that are immunocompromised, the high mortality rate associated with human listeriosis makes it the leading cause of death among foodborne bacterial pathogens. As a result, tremendous effort has been made to develop methods for the isolation, detection and control of L.monocytogenes in foods.

Salmonella

serotypes continue to be a prominent threat to food safety worldwide. Infections are commonly acquired by animal to human transmission though consumption of

undercooked food products derived from livestock or domestic fowl. The second half of the 20th century saw the emergence of
Salmonella serotypes that became associated with new food sources (i.e. chicken eggs) and the emergence of Salmonella serotypes with resistance against multiple antibiotics.

Shigella

species are members of the family Enterobacteriaceae and are Gram negative*, nonmotile rods. Symptoms include mild to severe diarrhea with or without blood, fever, tenesmus and abdominal pain. Further complications of the disease may be seizures, toxic megacolon, reactive arthritis and hemolytic uremic syndrome. Transmission of the pathogen is by the fecal-oral route, commonly through food and water. Shigella spp. are one of the leading causes of bacterial foodborne illnesses and can spread quickly within a population.

Escherichia coli

E. coli was considered a commensal of human and animal intestinal tracts with low virulence potential. It is now known that many strains of E. coli
act as pathogens, inducing serious gastrointestinal diseases and even death in humans.

Clostridium botulinum and Clostridium perfringens

produces extremely potent neurotoxins that result in the severe
neuroparalytic disease, botulism. The enterotoxin produced by C. perfringens during sporulation of vegetative cells in the host intestine results in debilitating acute diarrhea and abdominal pain. Sales of refrigerated, processed foods of extended durability including sous-vide foods, chilled ready-to-eat meals, and cook-chill foods have increased over recent years. Anaerobic spore-formers have been identified as the primary microbiological concerns in these foods.

Bacillus cereus

is a normal soil inhabitant, and is frequently isolated from a variety of foods, including vegetables, dairy products and meat. It causes a vomiting or diarrhea illness that is becoming increasingly important in the industrialized world. Some patients may experience both types of illness simultaneously. The diarrheal type of illness is most prevalent in the western hemisphere, whereas the emetic type is most prevalent in Japan. Desserts, meat dishes, and dairy products are the foods most frequently associated with diarrheal illness, whereas rice and pasta are the most common vehicles of emetic illness.

Prevention of Foodborne Illness

1)Cook-Cook all meat, poultry and eggs to at least 160F. Other than spore-forming bacteria, all bacteria, parasites and viruses are killed quite easily with heating to 160F.
2)Avoid Cross-Contamination-Do not cross-contaminate one food with another. Keep raw food totally separated from cooked product. Clean utensils and work areas etc in

between working raw and cooked product. Constantly be thinking of how microorganisms get from raw to cooked products.

3)Chill Foods-Keep foods cold. After cooking, chill foods as rapidly as possible. Remember that cooking has destroyed most of the bacteria but spore formers, that are resistant to cooking may become very active and can proliferate rapidly.

4)Cleaning-Wash fruits and vegetables and all foods possible. In addition, continually wash work areas. Use only treated or tested water.

5)Personal Hygiene-People working with foods should wash their hands regularly, wear hairnets, plastic gloves etc. In addition, food handlers should not work with food if they have a boil, open sores or feel sick themselves

Practical: Microbial food poisoning

Objectives

Recognize clinical manifestations of botulism, salmonella and staph food poisoning.
Outline the transmission, reservoirs and laboratory diagnostic approach in different microbial food poisoning.
Outline different preventive measures of microbial food poisoning.

Foodborne Botulism

It results from the ingestion of preformed toxin present in contaminated food. Symptoms usually develop between 12 and 36 hours after toxin ingestion. The initial complaints may be gastrointestinal and can include nausea, vomiting, abdominal cramps or diarrhea. Constipation is more likely to occur after the onset of neurologic symptoms. Dry mouth, blurred vision, and diplopia are the earliest neurologic symptoms. Lower cranial nerve dysfunction manifests as; dysphasia (difficulty speaking), dysarthria (difficulty articulating), and hypoglossal (tongue) weakness. Symmetric weakness then descends to the upper extremities, the trunk, and the lower extremities. Respiratory dysfunction may require ventilation. Autonomic problems may also include; alterations in resting heart rate, loss of responsiveness to hypotension or postural change, hypothermia, and urinary retention. Recovery may not begin for up to 100 days and may take months to conclude. With critical care management, the death rate is about 14%.

Reservoir

Botulism is caused by a nerve toxin, botulin, produced by the bacterium Clostridium(C) botulinum. Botulin is the most lethal substance known with less than one microgram sufficient to cause fatal human disease. C. botulinum, commonly found in soil, form spores which allow them to survive in a dormant state until exposed to conditions that can support their growth.

Transmission

There are four mechanisms for botulism toxin to enter the body: foodborne, cutaneous, colonization of the gastrointestinal tract, and inhalational. Foodborne botulism results when food contaminated with spores of

C. botulinum are ingested. Food contamination can occur if foods are preserved or stored under conditions that allow toxin production.

Diagnosis

Botulism diagnosis is based primarily on clinical presentation and should be suspected in a person with acute onset of gastrointestinal, autonomic (such as dry mouth or difficulty focusing eyes), and cranial-nerve dysfunction (diplopia, dysarthria, dysphagia). The diagnosis is even more likely if the patient has recently eaten home-canned foods or if family members/companions who have shared the same meals are similarly ill.

Treatment:

Persons with botulism require immediate emergency medical treatment. Treatment must not await laboratory confirmation.

•Botulism antitoxin and immune globulin

Preventive Measures

•Wash hands prior to and after preparing food

•Keep all work surfaces, food, utensils, equipment clean during all stages of food preparation especially for canning processes

•Date and label preserves and canned goods and follow proper canning requirements strictly

•Refrigerate all foods labeled "keep refrigerated"

•Methods to control botulism should focus on the inhibition of bacterial growth and toxin production. Manufacturers of commercially canned low acid foods use strict thermal processes which are designed to destroy spores of C. botulinum

•Educate the public about safe handling of food. For example: Do not use food from damaged or bulging containers.Foods with off-odours and unusual tastes should not be eaten.

Salmonella food poisoning:

Salmonellosis is caused by gram negative non-spore forming bacilli belonging to the Enterobactericeae family. There are more than 2460 serotypes. The most common serotypes that cause human disease are divided among the O

-antigen groups A through E. Salmonella serotype Typhimurium (serotype B) and E. Salmonella serotype Enteritidis (serotype D) are the most commonly reported human isolates

Clinical Presentation

Salmonella organisms can cause asymptomatic carriage, gastroenteritis, bacteremia and focal infections such as meningitis and osteomyelitis.

The most common illness associated with a Salmonella infection is gastroenteritis, in which diarrhea, abdominal cramps, and fever are common manifestations. The site of infection usually is the small intestine but colitis can occur. The illness usually lasts 4 to 7days and most people recover without treatment. The very young, the elderly and immunosuppressed persons are more at risk for complications.

Diagnosis

The diagnosis is made through the isolation of Salmonella from feces, rectal swabs or other body fluids. Freshly passed stool is preferred.

Reservoir

Salmonella species are widely present in animal reservoirs including poultry, birds, reptiles, livestock, rodents, pets; such as iguanas, tortoises, turtles, dogs and cats; also humans.

Transmission

The major mode of transmission is fecal oral route. (i.e. ingestion of food or water contaminated with animal or human feces). Another source of transmission is food of animal origin, such as poultry, beef, eggs, and dairy products. Other food vehicles (e.g., fruits, vegetables, and bakery products) have been implicated in outbreaks, in which the food was contaminated by contact with an infected animal product or human. Other modes of transmission include ingestion of contaminated water; contact with infected

reptiles or amphibians and possibly rodents; and exposure to contaminated medications, dyes, and medical instruments.

Incubation Period

Is from 6 to 72 hours, usually 12-36 hours.

Communicability

The risk of human-to-human transmission exists for the duration of fecal excretion of organisms.

Treatment

•Rehydration and electrolyte replacement should be provided.

•Antibiotic treatment may be recommended by a physician. Antibiotic therapy may be recommended for:

O individuals with severe disease,

o individuals with systemic illness including septicemia,

o the very young (< two months), elderly, and debilitated persons,

o individuals with cardiac valvular or endovascular abnormalities,

o persons with HIV, and

o individuals with other immunocompromised states.

•The use of antibiotics is often not effective in eradicating the carriage of Salmonella if anatomic abnormalities (i.e., biliary or kidney stones) are present.

Management of Contacts

•Symptomatic contacts must be referred for investigation. Investigation of contacts should include stool cultures of any household contacts who are involved in food handling, direct patient care, or care of young children or elderly people in institutional settings.

•Contacts should be given on information on disease transmission and appropriate personal hygiene.

Preventive Measures

•Educate the public about good sanitation and personal hygiene

•Food establishments should ensure compliance with the Food Premises Regulations.

•Follow the clean, separate, cook and chill rules

O Clean: wash hands and surfaces often

o Wash hands before and after handling food and after using the bathroom, changing diapers, and handling pets.

O Wash utensils, cutting boards, dishes, and countertops after preparing each food item and before you go on to the next item

O Separate: Don't cross-contaminate

O Separate raw meat, poultry, and seafood from other foods in the grocery shopping cart and in the refrigerator.

O Always wash cutting boards, dishes, countertops, and utensils after they come in contact with raw meat, poultry, and seafood.

O Never place cooked food on a plate that previously held raw meat, poultry, or seafood

O Cook: Cook to safe temperatures

O Use a clean food thermometer when measuring the internal temperature of meat, poultry, casseroles, and other foods to make sure they have reached a safe minimum internal temperature

O Do not eat or drink foods containing raw eggs, or unpasteurized milk

O Chill: Refrigerate promptly

O Keep food safe at home, refrigerate promptly and properly. Refrigerate or freeze perishables, prepared foods, and leftovers within 2 hours (1 hour if temperatures are above 90 °F)

O Freezers should register 0 °F or below and refrigerators 40 °F or below

O Thaw food in the refrigerator, in cold water, or in the microwave. Foods should not be thawed at room temperature

O Foods thawed in the microwave or in cold water must be cooked to a safe minimum internal temperature before refrigerating

o Marinate foods in the refrigerator

o Divide large amounts of leftovers into shallow containers for quick cooling in the refrigerator
o Don't pack the refrigerator. Cool air must circulate to keep food safe.

Staphylococcal food poisoning:

It is a self-limiting disease that occurs following ingestion of preformed **enterotoxin** in contaminated food. The organism reaches the food from the hands or nose of *S. aureus* shedders (***nasal carriers*** or ***cases***). The organisms multiply in the food and produce enterotoxin. The toxin does not change the taste, color, or odor of the food. Foods most commonly involved are briefly cooked and rich in carbohydrates as milk, milk products, cakes, pastry, koshari and koskosi.

Incubation period is short (1-6 hours) followed by severe symptoms: abdominal colic, nausea, **vomiting** (toxins directly act on the vomiting center) and diarrhea, but without fever.

Laboratory Diagnosis of S. aureus Food Poisoning:

The diagnosis is made through the isolation of S.aureus and detection of **Enterotoxin** from Remnants of food, vomitus or stool.

Chapter 12: Molecular genetics, epigenetics and nutrigenomics

Objectives

Discuss the concepts of epigenetics and nutrigenomics.
Recognize different techniques of molecular genetics.
Recognize the genetic screening, gene therapy.
Understand the principals of human genome project.

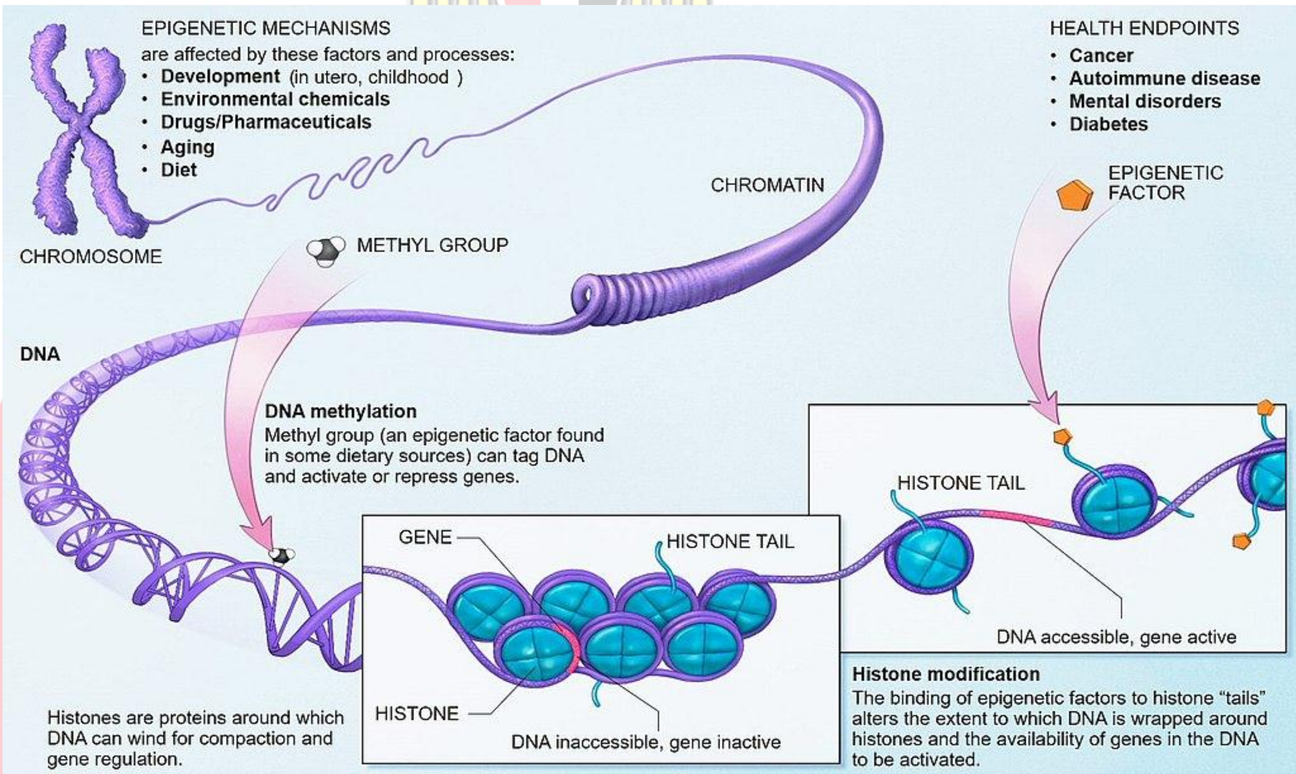
Overview of molecular genetics and related concepts

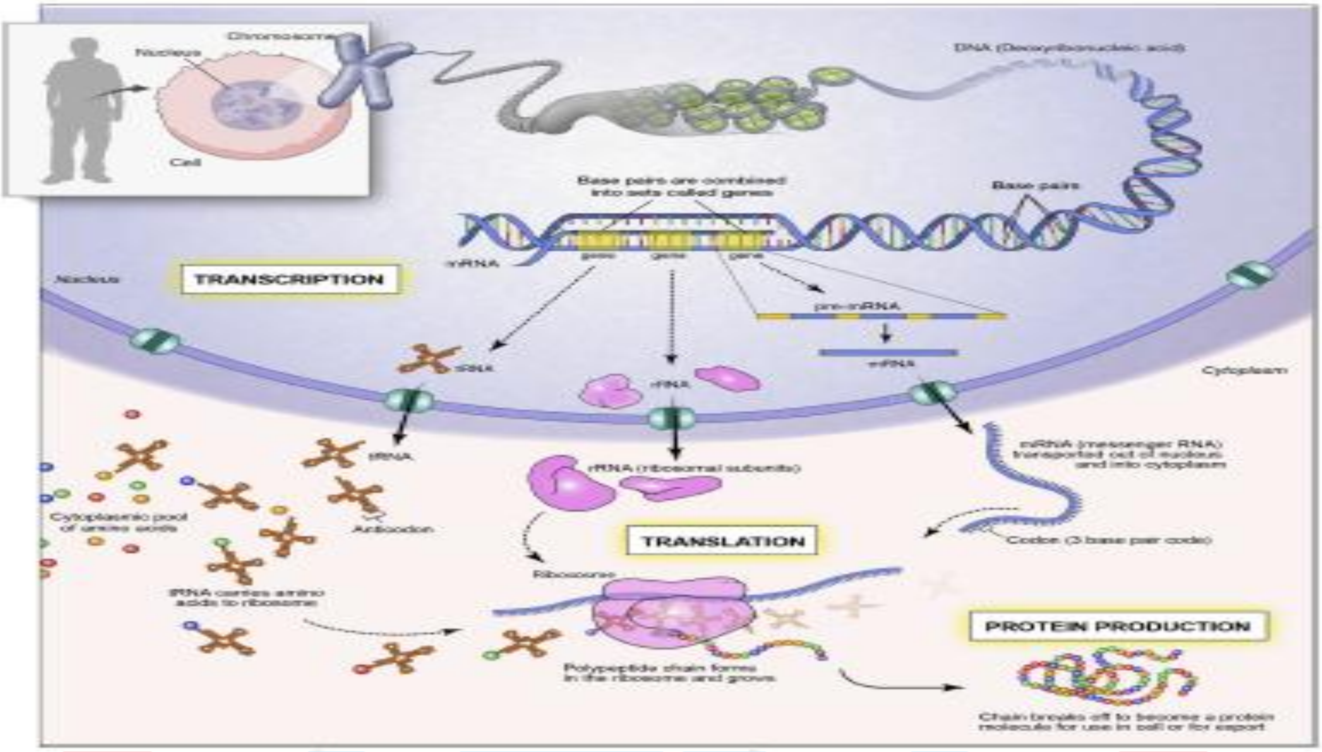
Molecular genetics is the field of biology that studies the structure and function of genes at a molecular level and thus employs methods of both molecular biology and genetics. The study of chromosomes and gene expression of an organism can give insight into heredity, genetic variation, and mutations. This is useful in the study of developmental biology and in understanding and treating genetic diseases.

Epigenetics is the study of heritable phenotype changes that do not involve alterations in the Deoxy nucleic acid (DNA) sequence. Epigenetics most often denotes changes that affect gene activity and expression, but can also be used to describe any heritable phenotypic change. Such effects on cellular and physiological phenotypic traits may result from external or environmental factors, or be part of normal developmental program. The standard definition of epigenetics requires these alterations to be heritable, either in the progeny of cells or of organisms.

The term also refers to the changes themselves: functionally relevant changes to the genome that do not involve a change in the nucleotide sequence. Examples of mechanisms that produce such changes are DNA methylation and histone modification, each of which alters how genes are expressed without altering the underlying DNA sequence. Gene expression can be controlled through the action of repressor proteins that attach to silencer regions of the DNA. These epigenetic changes may last through cell divisions for the duration of the cell's life, and may also last for multiple generations even though they do not involve changes in the underlying DNA sequence of the organism; instead, non-genetic factors cause the organism's genes to behave (or "express themselves") differently.

One example of an epigenetic change in eukaryotic biology is the process of cellular differentiation. During morphogenesis, totipotent stem cells become the various pluripotent cell lines of the embryo, which in turn become fully differentiated cells. In other words, as a single fertilized egg cell – the zygote – continues to divide, the resulting daughter cells change into all the different cell types in an organism, including neurons, muscle cells, epithelium, endothelium of blood vessels, etc.,by activating some genes while inhibiting the expression of others.





Nutritional genomics is a science studying the relationship between human genome, nutrition and health.

It can be divided into two disciplines:

- Nutrigenomics: studies the effect of nutrients on health through altering genome, proteome, metabolome and the resulting changes in physiology.
- Nutrigenetics: studies the effect of genetic variations on the interaction between diet and health with implications to susceptible subgroups. More specifically, nutrigenetics studies how individual differences in genes influence the body's response to diet and nutrition. For example, people with an enzyme deficiency caused by mutations in the enzyme phenylalanine hydroxylase cannot metabolize foods containing the amino acid phenylalanine and must modify their diets to minimize consumption. With modern genomic data, severe gene mutations with less severe effects are being explored to determine whether dietary practices can be more closely personalized to individual genetic profiles.

Nutrigenomics refers to the prospective analysis of differences among nutrients in the regulation of gene expression i.e., it studies the effect of nutrients on the genome, proteome, and metabolome. It involves the application of high-throughput genomic tools such as DNA microarray technology in nutrition research. Nutrigenomics is a discovery science which aims at understanding how nutrition influences metabolic pathways and homeostatic control and how this regulation is disturbed in the early phase of a diet-related disease.

Technique:

Amplification

Gene amplification is a procedure in which a certain gene or DNA sequence is replicated many times in a process called DNA replication.

Polymerase chain reaction

The main genetic components of the polymerase chain reaction (PCR) are DNA nucleotides, template DNA, primers and Taq polymerase. DNA nucleotides make up the DNA template strand for the specific sequence being amplified and primers are short strands of complementary nucleotides where DNA replication starts. Taq polymerase is a heat stable enzyme that jump-starts the production of new DNA at the high temperatures needed for reaction.

Cloning DNA in bacteria

Cloning is the process of creating many identical copies of a sequence of DNA. The target DNA sequence is inserted into a cloning vector. Because this vector originates from a self-replicating virus, plasmid, or higher organism cell, when the appropriate size DNA is inserted, the "target and vector DNA fragments are then ligated" to create a recombinant DNA molecule.

The recombinant DNA molecule is then injected into a bacterial strain (usually *E. coli*) which produces several identical copies of the selected sequence it absorbed through transformation (the mechanism by which bacteria uptake foreign DNA from the environment into their genomes).

Separation and detection

In separation and detection, DNA and mRNA are isolated from cells and then detected simply by the isolation. Cell cultures are also grown to provide a constant supply of cells ready for isolation.

Cell cultures

A cell culture for molecular genetics is a culture that is grown in artificial conditions. Some cell types grow well in cultures such as skin cells, but other cells are not as productive in cultures. There are different techniques for each type of cell, some only recently being found to foster growth in stem and nerve cells. Cultures for molecular genetics are frozen in order to preserve all copies of the gene specimen and thawed only when needed. This allows for a steady supply of cells.

DNA isolation

DNA isolation extracts DNA from a cell in a pure form. First, the DNA is separated from cellular components such as proteins, RNA, and lipids. This is done by placing the chosen cells in a tube with a solution that mechanically, chemically, breaks the cells open. This solution contains enzymes, chemicals, and salts that breaks down the cells except for the DNA. It contains enzymes to dissolve proteins, chemicals to destroy all RNA present, and salts to help pull DNA out of the solution. Next, the DNA is separated from the solution by being spun in a centrifuge, which allows the DNA to collect in the bottom of the tube. After this cycle in the centrifuge the solution is poured off and the DNA is resuspended in a second solution that makes the DNA easy to work with in the future. This results in a concentrated DNA sample that contains thousands of copies of each gene. For large scale projects such as sequencing the human genome.

mRNA isolation

Expressed DNA that codes for the synthesis of a protein is the final goal for scientists and this expressed DNA is obtained by isolating mRNA (Messenger RNA).

First, laboratories use a normal cellular modification of mRNA that adds up to 200 adenine nucleotides to the end of the molecule (poly(A) tail). Once this has been added, the cell is ruptured and its cell contents are exposed to synthetic beads that are coated with thymine string nucleotides. Because Adenine and Thymine pair together in DNA, the poly(A) tail and synthetic beads are attracted to one another, and once they bind in this process the cell components can be washed away without removing the mRNA. Once the mRNA has been isolated, reverse transcriptase is employed to convert it to single-stranded DNA, from which a stable double-stranded DNA is produced using DNA polymerase. Complementary DNA (cDNA) is much more stable than mRNA and so, once the double-stranded DNA has been produced it represents the expressed DNA sequence scientists look for.

Genetic screens

Forward genetics

This technique is used to identify which genes or genetic mutations produce a certain phenotype. A mutagen is very often used to accelerate this process. Once mutants have been isolated, the mutated genes can be molecularly identified. Forward saturation genetics is a method for treating organisms with a mutagen, then screens the organism's offspring for particular phenotypes. This type of genetic screening is used to find and identify all the genes involved in a trait.

Reverse genetics

Reverse genetics determines the phenotype that results from a specifically engineered gene. In some organisms, such as yeast and mice, it is possible to induce the deletion of a particular gene, creating what's known as a gene "knockout" - the laboratory origin of so-called "knockout mice" for further study. In other words this process involves the creation of transgenic organisms that do not express a gene of interest. Alternative methods of reverse genetic research include the random induction of DNA deletions and subsequent selection for deletions in a gene of interest, as well as the application of Ribonucleic acid (RNA) interference.

Gene therapy

A mutation in a gene can cause encoded proteins and the cells that rely on those proteins to malfunction. Conditions related to gene mutations are called genetic disorders. However, altering a patient's genes can sometimes be used to treat or cure a disease as well. Gene therapy can be used to replace a mutated gene with the correct copy of the gene, to inactivate or knockout the expression of a malfunctioning gene, or to introduce a foreign gene to the body to help fight disease. Major diseases that can be treated with gene therapy include viral infections, cancers, and inherited disorders, including immune system disorders.

Gene therapy delivers a copy of the missing, mutated, or desired gene via a modified virus or vector to the patient's target cells so that a functional form of the protein can then be produced and incorporated into the body. These vectors are often siRNA. Treatment can be either in vivo or ex vivo. The therapy has to be repeated several times for the infected patient to continually be relieved, as repeated cell division and cell death slowly randomizes the body's ratio of functional-to-mutant genes. Gene therapy is an appealing alternative to some drug-based approaches, because gene therapy repairs the underlying genetic defect using the patients own cells with minimal side effects. Gene therapies are still in development and mostly used in research settings.

Classical gene therapies usually require efficient transfer of cloned genes into the disease cells so that the introduced genes are expressed at sufficiently high levels to change the patient's physiology. There are several different physicochemical and biological methods that can be used to transfer genes into human cells. The size of the DNA fragments that can be transferred is very limited, and often the transferred gene is not a conventional gene. Horizontal gene transfer is the transfer of genetic material from one cell to another that is not its offspring. Artificial horizontal gene transfer is a form of genetic engineering.

The Human Genome Project

The Human Genome Project is a molecular genetics project that began in the 1990s and was projected to take fifteen years to complete. However, because of technological advances the progress of the project was advanced and the project finished in 2003, taking only thirteen years. The project was started by the U.S. Department of Energy and the National Institutes of Health in an effort to reach six set goals. These goals included:

1. identifying 20,000 to 25,000 genes in human DNA (although initial estimates were approximately 100,000 genes),
2. determining sequences of chemical base pairs in human DNA,
3. storing all found information into databases,
4. improving the tools used for data analysis,
5. transferring technologies to private sectors, and
6. addressing the ethical, legal, and social issues that may arise from the projects.

The project was worked on by eighteen different countries including the United States, Japan, France, Germany, and the United Kingdom. The collaborative effort resulted in the discovery of the many benefits of molecular genetics. Discoveries such as molecular medicine, new energy sources and environmental applications, DNA forensics, and livestock breeding, are only a few of the benefits that molecular genetics can provide.

Biomics technologies

The recent advances in nutrigenomics studies are owed to the completion of human genome project and the new biomics technologies that provide means for the simultaneous determination of the expression of many thousands of genes at the mRNA (transcriptomics), metabolites (metabolomics) and protein (proteomics) levels. Genomic and transcriptomic studies are mostly conducted by DNA microarray technologies. Proteomics and metabolomics have no standardized procedures yet, but usually, proteome analysis is done by two-dimensional gel electrophoresis and Liquid chromatography-mass spectrometry, while metabolome analysis is conducted through gas chromatography-mass spectrometry, liquid chromatography-mass spectrometry and liquid chromatography-nuclear magnetic resonance. Usually, these technologies are applied in a "differential display" mode, i.e. by comparing two situations (e.g. diseased versus healthy) in order to reduce the complexity in data by examining only differences.

PERSONALIZED NUTRITION

Personalized nutrition uses familial, genetic, or metabolomics information to interpret an individual's health risk profile. The derived nutritional recommendations are claimed to help maintain wellness and/or reduce disease risk.

A human phenotype is the composite of observable characteristics or traits, including appearance, behavior, development, and biochemical or physiological properties. Phenotypes result from the interaction between genes and environment, which ultimately determines the personalized nutritional requirements for an individual.

There are a considerable number of genes that affect individual dietary requirements. However, we are not yet sufficiently in control of bioinformatics manipulation of that genetic information to understand how to optimally combine information on gene pathways and epistasis, thereby determining individual nutritional requirements.

The identification of responders from non responders to diet must be a primary goal of personalizing nutrition, based on genetic and metabolic information. The response of an individual to the combined effects of nutrient and caloric intake, genetic and epigenetic background, lifestyle choices, and environmental exposures, provides a sensitive indicator of nutritional and metabolic status, increasingly being measured as a metabolic phenotype. Such information enables a rational basis for the selection of foods, including functional foods, and supplements, along with lifestyle modification, to move an individual's health in a more personally beneficial direction. The ultimate goal is to develop a dietary pattern for each individual to maximize health and wellness, and prevent disease. This will also depend on age, activity level, and other lifestyle and environmental factors.

Practical: The role of dietetic therapy, screening, diagnosis and genetic counseling of genetic and metabolic disorders

Objectives

Understand Gene-diet-disease interaction
Recognize the role of Nutrigenetics in Type 2 Diabetes mellitus
Recognize the role of Nutrigenetics in cardiovascular diseases.
Recognize the role of Nutrigenetics in cancer.

Gene-diet-disease interaction

97% of the genes known to be associated with human diseases result in *monogenic diseases*, i.e. a mutation in one gene is sufficient to cause the disease. Modifying the dietary intake can prevent some monogenic diseases. One example is phenylketonuria, a genetic disease characterized by a defective phenylalanine hydroxylase enzyme, which is normally responsible for the metabolism of phenylalanine to tyrosine. This results in the accumulation of phenylalanine and its breakdown products in the blood and the decrease in tyrosine, which increases the risk of neurological damage and intellectual disabilities. Phenylalanine-restricted tyrosine-supplemented diets are a means to nutritionally treat this monogenic disease.

In contrast, many common diseases, such as obesity, cancer, diabetes, and cardiovascular diseases, are *polygenic diseases*, i.e. they arise from the dysfunction in a cascade of genes, and not from a single mutated gene. Dietary intervention to prevent the onset of such diseases is a complex and ambitious goal.

Recently, it was discovered that the health effects of food compounds are related mostly to specific interactions on molecular level, i.e. dietary constituents participate in the regulation of gene expression by modulating the activity of transcription factors, or through the secretion of hormones that in turn interfere with a transcription factor.

A number of genetic variations have been shown to increase the susceptibility to diet-related diseases. These include variants that have been associated with Type 2 diabetes mellitus, obesity, cardiovascular diseases, some autoimmune diseases and cancers. Nutrigenetics aims to study these susceptible genes and provide dietary interventions for individuals at risk of such diseases. Some examples are shown below.

Applications of nutrigenetics:

A number of genetic variations have been shown to increase the susceptibility to diet-related diseases. These include variants that have been associated with Type 2 diabetes mellitus, obesity, cardiovascular diseases, some autoimmune diseases and cancers. Nutrigenetics aims to study these susceptible genes and provide dietary interventions for individuals at risk of such diseases.

Nutrigenetics and Type 2 Diabetes mellitus

A number of genes are involved in regulating lipid metabolism and insulin sensitivity, and thereby affecting the susceptibility to type 2 diabetes mellitus. Among them is the gene responsible for sterol response element binding protein-1c or SREBP-1c (a membrane-bound transcription factor which can directly activate the expression of several genes involved in the synthesis and uptake of cholesterol, fatty acids, triglycerides and phospholipids).

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Nutrigenetics and cardiovascular diseases

Hyperlipidemia is usually associated with atherosclerosis and coronary heart disease. Therapy includes lifestyle changes as alterations in the patient's diet, physical activity and treatment with pharmaceuticals such as statins. However, individuals respond differently to the treatment. This was attributed to genetic variations within the population. Genetic variations in genes encoding for apolipoproteins, some enzymes and hormones can alter individual sensitivity to developing cardiovascular diseases. Some of these variants are susceptible for dietary intervention, for example:

- Individuals with the E4 allele in the apolipoprotein E gene show higher low-density lipoprotein-cholesterol (bad cholesterol) levels with increased dietary fat intake compared with those with the other (E1, E2, E3) alleles receiving equivalent amounts of dietary fat.
- One single nucleotide polymorphism (-75 G/A) in the apolipoprotein A1 gene in women is associated with an increase in High density lipoprotein-cholesterol levels with the increase in the dietary intake of polyunsaturated fatty acids. Individuals with the A variant showed an increase in the protective HDL (good cholesterol) levels following an increased consumption of PUFA compared with those with the G variant taking similar amounts of PUFA.

- One polymorphism (-514 CC) in the hepatic lipase gene is associated with an increase in protective HDL levels compared with the TT genotype (common in certain ethnic groups such as African-Americans) in response to high fat diet.

Nutrigenetics and cancer

Nutrients can contribute to the development of cancers especially colon, gastric and breast cancer. Several gene variants have been identified as susceptibility genes. One example is the N-Acetyltransferase (NAT) gene. NAT is a metabolism enzyme that exists in two forms: NAT1 and NAT2. Several polymorphisms exist in NAT1 and NAT2. NAT is involved in acetylation of heterocyclic aromatic amines found in heated products especially **well cooked red meat**. During cooking of muscle meat at high temperature, some amino acids may react with creatine to give heterocyclic aromatic amines (HAA). HAA can be activated through acetylation to reactive metabolites which bind **DNA and cause cancers**. Only NAT2 fast acetylators can perform this acetylation. So NAT2 fast acetylator genotype had a higher risk of developing colon cancer in people who consumed relatively large quantities of red meat.

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