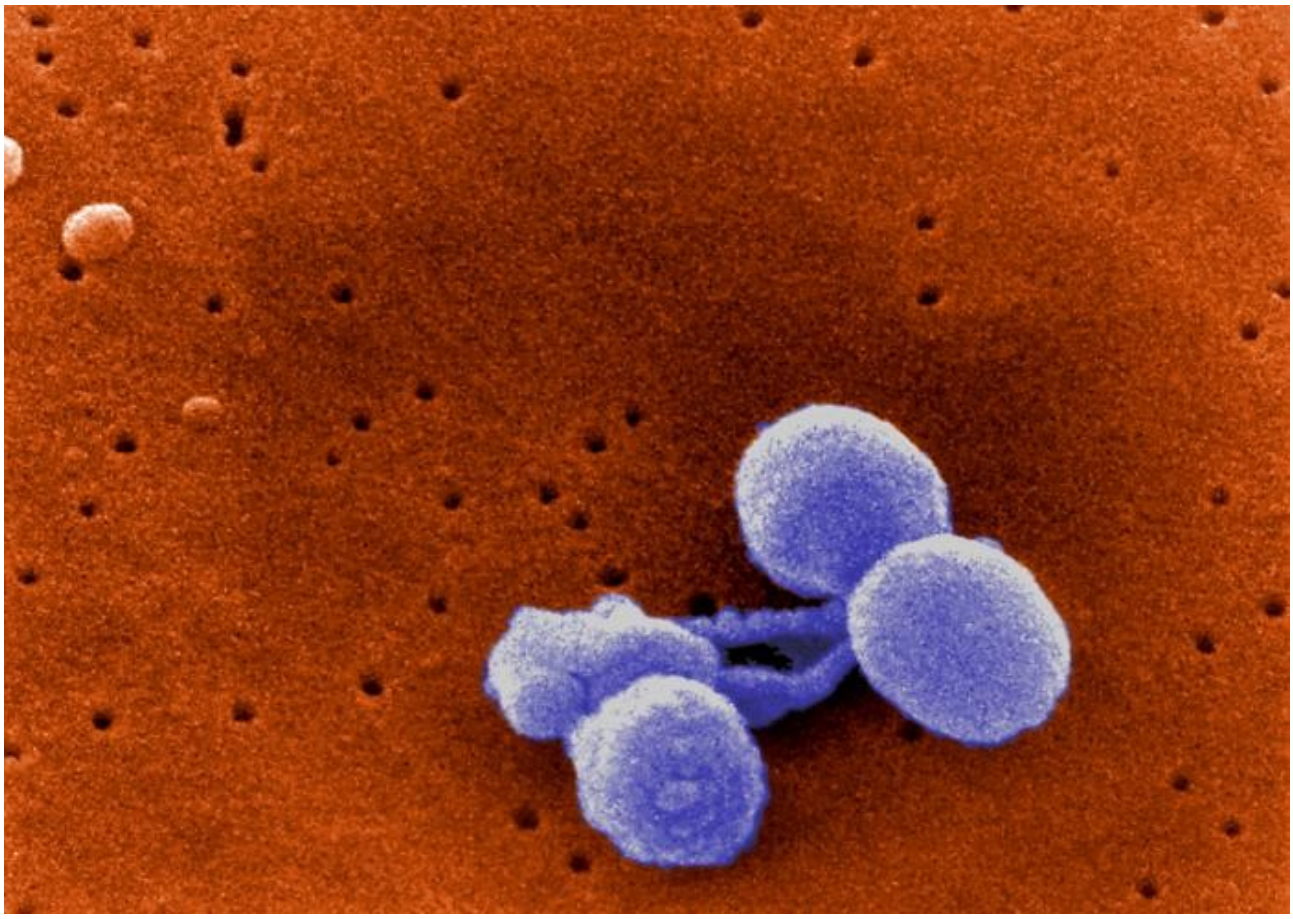
Meningitis:

Primary causative agents and epidemiological study of diagnosed cases of bacterial meningitis in Catalonia from 1994 to 2009



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1. Introduction

When the idea of conducting a research project on meningitis first occurred to me, it was because of my interest in microbiology and my intention to continue my studies in health sciences. I wanted my project to reflect my field of interest.

Microbiology is a wide field of expertise and selecting one disease from it, though difficult, seemed to be the most favourable choice of research, as it also involved my interest in infectious diseases. I did have parameters with which to select this disease in that I wanted to choose something that could be encountered in the community I live in.

Meningitis was far from eradicated in western nations, as was made evident by the still-present public concern for the infection and so, it made the disease a far more relevant investigation subject than, say, the Ebola virus.

I knew nothing about the transmission mechanisms of the disease, where it struck or even what constituted the meninges. In addition, I could not find any research project done on the topic of meningitis before, something which I felt added to the originality of the research.

Public sensitivity towards the disease (meningitis was, and still is, a cause for serious public concern), promoted by the heightened sequelae and mortality rates, also made the prospective research seem that much more valuable.

When I did some basic information-gathering on the short list of conditions I had accumulated and discovered that meningitis had potential causes in several of the most important subfields of microbiology (bacterial, viral and fungal infections), I decided to put it forward as my primary research topic selection.

2. Anatomy of the meninges

Note: unless otherwise specified, the sources for the anatomy of the meninges section are the 20th and 40th editions of Henry Gray's Anatomy of the Human Body.

The meninges is the system of membranes that covers the **central nervous system**. The central nervous system is contained within the dorsal cavity, and contains the brain, within the cranial cavity, and the spinal cord, within the spinal cavity. The composition and description of the meninges often varies between those portions which are positioned within the cranial cavity and those positioned within the spinal cavity.

The meninges consists of three different layers known as the **dura mater**, the **arachnoid mater**, and the **pia mater**. The primary function of these membranes is protection.

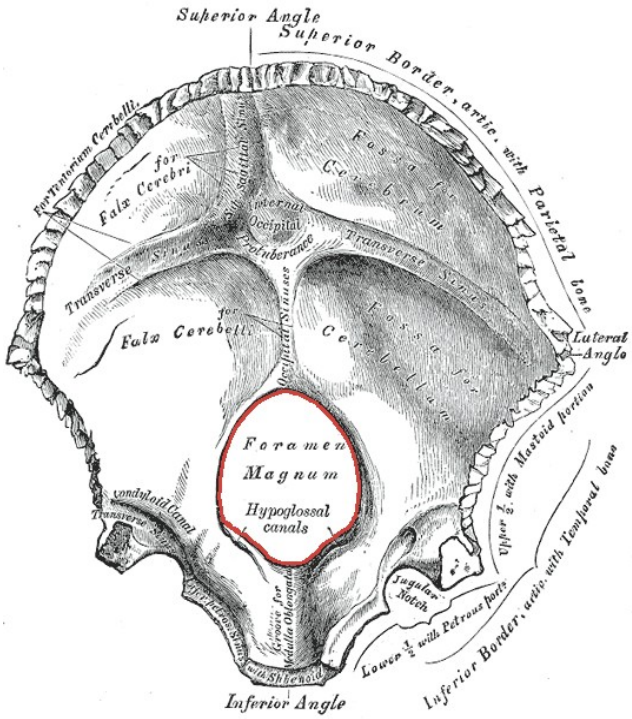

fig. 1 The foramen magnus is a large opening in the occipital bone of the cranium, through which the spinal cord enters and exits the skull. Pictured also is the falx cerebri (upper-left process) and the falx cerebelli (lower-left process). *Adapted from Henry Gray's "Anatomy of the Human Body", 20th edition.*

2.1 Dura mater

The **dura mater** is thick, dense and inelastic, and the most external membrane of the meningeal system. The dura mater can be described in two portions that together make up the complete membrane and are constant at the foramen magnum[1], the portion of the mater that encloses the brain and the portion that surrounds the medulla spinalis or spinal cord:

The **cranial dura mater** lines the inside of the skull, and serves two functions: functioning as

1 Foramen magnum: An opening in the skull where cranial dura becomes known as spiral dura (see fig. 1).

the internal periosteum[2] of the cranium and as a membrane of protection for the brain. This area of the dura mater is composed of two layers, the inner or meningeal layer and outer or endosteal layer. These two layers are closely connected, except when they separate to form cavities to allow the passage of deoxygenated blood. The outer surface of the cranial dura mater is rough, adhering closely to the inner cranium, whilst the inner surface is smooth and lined by a layer of endothelium (blood vessel-lining cell layer). The cranial dura mater is continuous with the spinal dura mater.

The cranial dura mater presents several folds of its inner, meningeal layer, known as **processes**[3], which project inward into the cavity of the skull. There are four processes: the falx cerebri (pictured in fig. 1 and fig. 2), the tentorium cerebelli (pictured in fig. 2), the falx cerebelli (pictured in fig. 1), and the diaphragma sellæ (a small circular fold, not pictured). These processes serve the function of separating various portions of the brain. The falx cerebri process is worthy of note, as it is the process which descends vertically in the medial longitudinal fissure[4], separating the cerebral hemispheres.

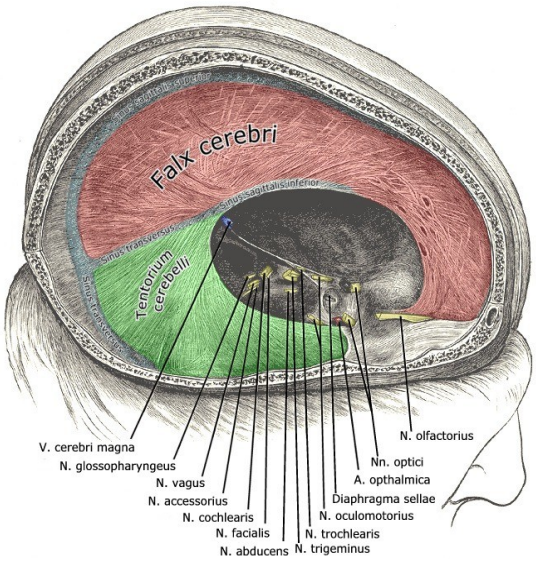

fig. 2 Pictured are the processes falx cerebri (red) and tentorium cerebelli (green). Adapted from Henry Gray's "Anatomy of the Human Body", 20th edition.

The **structure** of cranial dura mater consists of white fibrous tissues and elastic fibres arranged into various laminas. The outer, endosteal layer, which serves as an internal periosteum, contains the blood-vessels which supply the cranial bones. The inner, meningeal is lined with a layer of mesothelium[5]. The dura mater presents numerous arteries and veins which enter and exit the skull.

2 Periosteum: Connective tissue which provides nourishment to the bone it is adhered to.
3 Process: Outgrowth of tissue. In the context of the meninges, processes are folded protrusions found in the cranial cavity.
4 Medial longitudinal fissure: Deep groove that separates the two hemispheres of the brain.
5 Mesothelium: Cavity-lining membrane of connective tissue which provides a lubricating fluid which induces a slippery (and, therefore, non-adhesive), protective surface.

The **spinal dura mater** forms a loose sheath around the spinal cord, and presents only an inner, meningeal layer, as the function of the outer, endosteal layer is performed by the periosteum of the vertebral canal[6] at the foramen magnum. Although the spinal dura mater and the arachnoid mater are kept in contact, except when minute quantity of fluid separates them, the potential space between them is known as the **subdural cavity**. The dura mater is separated from the wall of the vertebral canal by the **epidural space**. The dura mater presents dual openings along the length of the spinal cord to allow the passage of spinal nerves, which the dura mater follows in the form of tubular prolongations, which, in turn, gradually lengthening from the upper part of the vertebral column to the lower spinal nerves below. The dura mater follows the spinal cavity until it blends with the periosteum of the coccyx.

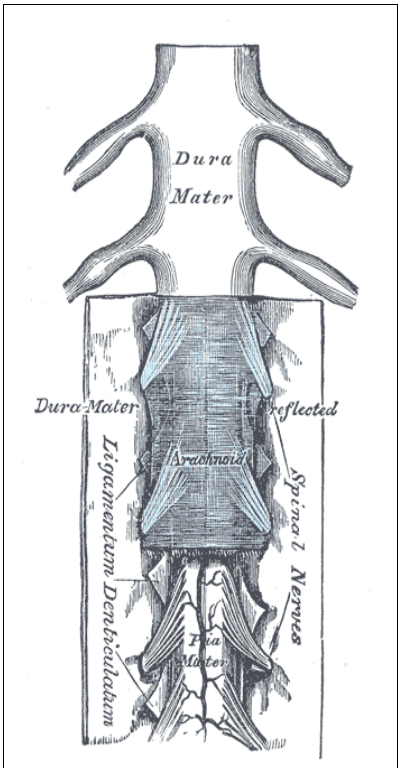

fig. 3 The spinal cord and its membranes, with double openings and prolongations visible along the dura mater to allow for the passage of spinal nerve roots. *Source: Henry Gray's "Anatomy of the Human Body", 20th edition.*

The **structure** of the spinal dura resembles the inner or meningeal layer of the cranial dura mater, and consists of white fibrous and elastic tissue. Its internal surface is smooth and is covered by a layer of mesothelium. It presents a moderate quantity of blood-vessels and a few nerves.

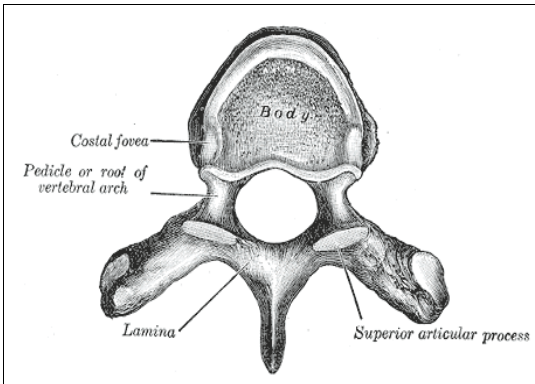

fig. 4 The space within the vertebrae known as the vertebral canal is pictured above. *Source: Henry Gray's "Anatomy of the Human Body", 20th edition.*

2.2 Arachnoid mater

The **arachnoid** is a delicate membrane which envelopes the brain and spinal cord, residing between the dura mater and the pia mater. Whilst it is usually constant with the external dura mater, it is completely separated from the

6 Vertebral canal: Intravertebral space which allows for the passage of the spinal cord (see fig. 4).

inner pia mater by the **subarachnoid cavity**, which is filled with **cerebrospinal fluid**. The arachnoid mater can be described in two segments, the segment which surrounds the brain, and the segment which surrounds the spinal cord.

The **cranial part** of the arachnoid mater surrounds the brain, without dipping into any sulci[7] between the gyri. The arachnoid membrane is thin and transparent on the upper surface of the brain, though becomes thicker and more opaque around the base and central parts of the brain.

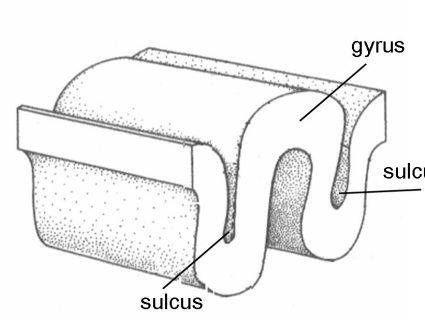

fig. 5 Pictured above are the sulci fissures that surround gyri on the surface of the cerebral cortex. Source: Henry Gray's "Anatomy of the Human Body", 20th edition.

The **spinal part** of the arachnoid mater surrounds the spinal cord. The spinal arachnoid is a thin, delicate and tubular membrane, and is continuous with the cranial arachnoid. It also widens out as it progresses down the spinal cord from the cranial arachnoid. The spinal arachnoid is separated from the spinal dura by the **subdural space**, although it maintains isolated beams connective tissue which traverse the space.

The **structure** of the arachnoid mater consists of white fibrous and elastic tissues. Its outer and inner surfaces are covered with a layer of mesothelium. Several large blood vessels are present in the structure of the arachnoid. The subarachnoid cavity is occupied by a spongy tissue consisting of delicate connective tissue, known as subarachnoid tissue. Whilst this cavity is small on the surface of the hemispheres of the brain, the arachnoid mater separates from the pia mater further along the sulci in between gyri (as the pia mater dips into the sulci, whereas the arachnoid mater bridges across from gyrus to gyrus). At certain parts of the base of the brain, the separation between arachnoid and pia maters is that of wide intervals which contain lessened quantities of subarachnoid tissue and are communicated freely with each, known as **subarachnoid cisternæ**. Protrusions of the arachnoid mater through the dura mater exist in the form of arachnoid granulations, which are one-way valves that serve to regulate pressure in the form of an exit for cerebrospinal fluid.

7 Sulci and gyri: Sulci are fissures that surround a gyrus, a ridge present on the cerebral cortex (see fig. 5)

The **cerebrospinal fluid system** is a protective system surrounding the brain and spinal cord on all sides, and is composed by the subarachnoid space and four cavities inside the brain (left and right lateral ventricles and third and fourth ventricles), each filled with cerebrospinal fluid and connected by a series of channels through which cerebrospinal fluid can flow. Cerebrospinal fluid is secreted by **choroid plexuses**, networks of blood vessels that protrude into each of the four cavities of the cerebrospinal fluid system inside the brain, secreting cerebrospinal fluid. Cerebrospinal fluid flows in one direction, from the lateral ventricles into the third ventricle, then to the fourth ventricle and finally into the subarachnoid space. Choroid plexuses act as direct blood-brain barriers, and, therefore, as potential routes of central nervous system infection for pathogens that have gained access to the bloodstream (Additional source: Anatomy and Physiology).

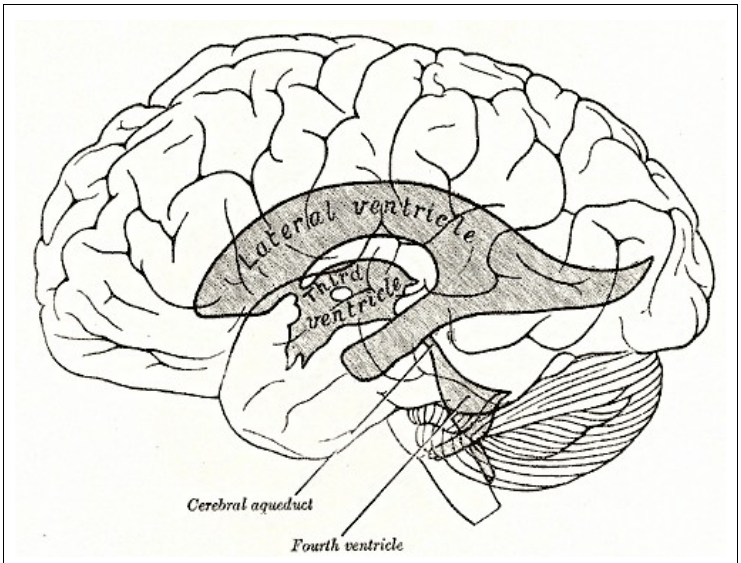

fig. 6 Image depicting the four cerebrospinal fluid system ventricles in the brain. The two lateral ventricles are viewed as one from the left. Source: Henry Gray's "Anatomy of the Human Body", 20th edition.

2.3 Pia mater

The **pia mater** is a highly vascular membrane held together by an extremely fine areolar tissue[8] covered by a layer of mesothelium projected from the inner arachnoid mater. The pia mater is an incomplete membrane, discontinuous at various openings to the general ventricular cavity of the brain and perforated by all blood-vessels that enter and leave the nervous system. Like the dura and arachnoid mater, the pia mater presents two segments, each continuous with the other.

8 Areolar tissue: Flexible, cushioning tissue of loosely organized fibres.

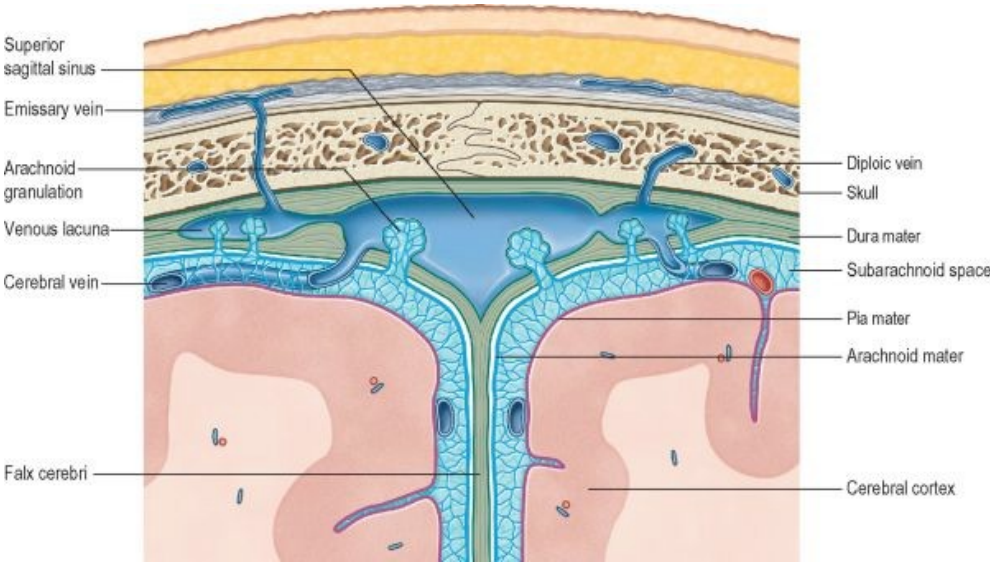

fig. 7 Meninges pictured above. Note the arachnoid granulation protruding into the superior sagittal sinus, a channel within the dura mater with the function of receiving blood from internal and external veins of the brain and cerebrospinal fluid from the subarachnoid space. Adapted from Henry Gray's "Anatomy of the Human Body", 40th edition.

The **cranial pia mater** surrounds the entire surface of the brain, dipping into sulci surrounding the various cerebral gyri.

The **spinal pia mater** is thicker and firmer than the cranial pia mater due to it presenting an additional layer of connective-tissue fibres. The spinal pia mater is intimately adhered to the spinal cord and spinal nerves, following the spinal cavity until it blends with the coccyx periosteum.

3. Prime causative agents of meningitis

3.1 Prime causative agents of bacterial meningitis

Meningitis can be defined as the inflammation of the arachnoid mater, pia mater and the intervening cerebrospinal fluid. Bacteria are the most prevalent cause of meningitis following viruses, and meningitis induced by bacteria is known as bacterial meningitis. It is usually far more severe than meningitis caused by viruses. There are over 50 types of bacteria capable of causing bacterial meningitis, of which the major types are detailed here (sources: Meningitis Research Foundation, Cecil Medicine).

3.1.1 *Neisseria meningitidis*

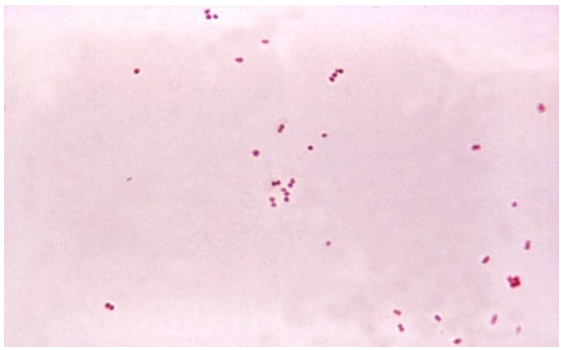

fig.1 Micrograph of the bacterium Neisseria meningitidis. Note the diplococcal group-arrangement. Source: Centers for Disease Control and Prevention.

Neisseria meningitidis (abbreviated *N. meningitidis*), also known as meningococcus, is a gram-negative[9], aerobic[10], encapsulated[11], non-spore forming, spherical bacterium with a diplococcus[12] group-arrangement. Infections caused by *N. meningitidis* are described as meningococcal disease. *N. meningitidis* is endemic to and exists as part of the normal flora in the nasopharynx[13] (in a non-pathogenic form) in approximately 10% of the human population, and is incapable of surviving outside the human body, being only transmissible from person to person through prolonged close contact, through the inhalation of aerosolised respiratory droplets. Meningococcal disease occurs when the bacteria break through the mucosal epithelium of the throat and nose of their host and enter the bloodstream (causing bacteremia[14]), where they multiply rapidly. *N. Meningitidis*' external capsule prevents the bacterium from phagocytosis[15] by phagocytes[16]. Once in the bloodstream, the bacteria can cross the blood-brain barrier[17] and enter the subarachnoid cavity, where they multiply freely in the cerebrospinal fluid, releasing endotoxic lipooligosaccharides[18], attracting an immune response and inflaming the meninges, causing meningitis (sources: Meningitis Research Foundation,

9 Gram-negative: Possible result of a Gram staining, an empirical method for determining whether a bacterium has a high concentration of peptidoglycan in its cell wall (retaining a purple colouration from the stain and being classified Gram-positive) or a low concentration (retaining a pink-red colouration and being classified Gram-negative).

10 Aerobic: Metabolism in which the respiration of oxygen is required, in contrast to an anaerobic metabolism.

11 Encapsulated: Property of bacteria that have an outer covering or "capsule" made of polysaccharide.

12 Diplococcus: Group-arrangement of cocci, which are spherically-shaped bacteria, in which the bacteria are arranged in two-cell pairs. Other group-arrangements include coccus (single bacteria) and streptococcus (chains of bacteria), bacillus (rod-shaped bacteria), coccobacillus (round-edged bacillus). See fig. 2.

13 Nasopharynx: Part of the throat situated immediately posterior to the mouth and nasal cavity.

14 Bacteremia: Presence of pathogenic bacteria in the bloodstream. Compare with septicemia, which also indicates the presence of bacteria in the blood, but is more often associated with severe infection.

15 Phagocytosis: Cellular process of incorporating a foreign particle of a volume superior to 150nm via the extension of a pseudopod, a temporary projection of the cytoplasm.

16 Phagocyte: Immune system cell that engulfs (via phagocytosis) and destroys invading viruses, bacteria and other pathogens.

17 Blood-brain barrier: Separation of circulating blood and brain extracellular fluid in the central nervous system.

18 Endotoxic lipooligosaccharides: Potent toxin that exists as part of the bacterial cell wall.

Sherris Medical Microbiology).

The result of these toxins being shed in the bloodstream is the meningococcal condition commonly associated with meningococcal meningitis known as meningococcemia, a subtype of septicemia (sources: Meningitis Research Foundation; University of California School of Medicine, PubMed).

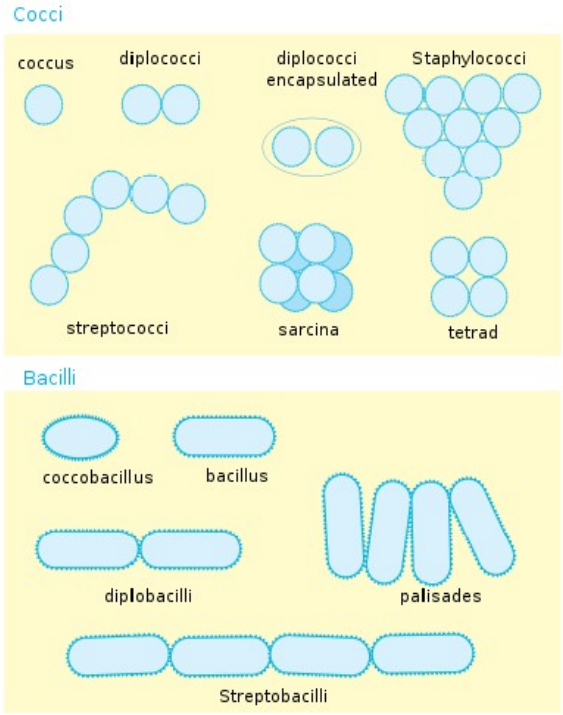

fig 2. Pictured above are several bacterial morphological group-arrangement. Source: Wikimedia Commons.

There are 12 serotypes[19] of *N. meningitidis*, of which 5 (A, B, C, Y and W-135) account for virtually all cases of meningococcal disease, with A, B and C being responsible for 90% of these cases, and 5 (A, B, C, W135 and X) account for the only strains of bacteria capable of causing epidemics of bacterial meningitis (sources: Institute for Clinical and Experimental Pathology, PubMed, Eurosurveillance, Norwegian Institute of Public Health).

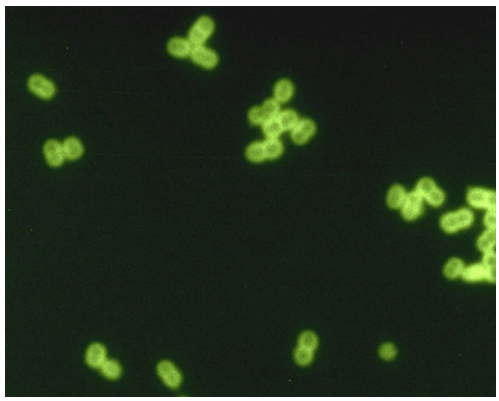

fig.3 Digitally-colourised fluorescent antibody-stain of the bacterium Streptococcus pneumoniae in spinal fluid. Note the varying group-arrangements. Source: Centers for Disease Control and Prevention.

3.1.2 *Streptococcus pneumoniae*

Streptococcus pneumoniae (abbreviated *S. pneumoniae*), also known as pneumococcus, is a Gram-positive, aerotolerant, anaerobic, non-spore forming, alpha-hemolytic[20], encapsulated, spherical, bacterium with a group-arrangement that varies between coccus, diplococcus and

19 Serotype: Collection of microorganisms of a species with a specific set of antigens (substances that induce immune responses). Contrast with strains, which are individual races of microbes, which, if antigenic, will belong to specific serotypes.

20 Alpha-hemolytic: Possible classification of individual species pertaining to the streptococcus genus based on the hemolytic (capacity to damage and rupture red blood cells) properties of each species when grown on blood agar, in which alpha-hemolysis indicates the oxidization of hemoglobin, beta-hemolysis indicates the

streptococcus. The bacteria exist in colonies in the normal flora of the nasopharyngeal tract, similarly to *N. meningitidis*, in 5-40% of the human population, with approximately half of children of pre-school age acting as carriers (sources: Meningitis Research Foundation, Sherris Medical Microbiology).

As with *N. meningitidis*, *S. pneumoniae* is transmissible only through close contact. Pneumococcal meningitis occurs when the bacteria break through the mucosal epithelium of the nasopharynx or other sites of infection, in the same manner as *N. Meningitidis* (gaining protection from phagocytes from its external capsule) and enter the bloodstream, spreading to the meninges and replicating in the cerebrospinal fluid beneath the arachnoid mater, with released toxins inducing meningeal inflammation. *S. pneumoniae* is also the causative agent of respiratory tract infections if it takes hold in the lungs and, like *N. meningitidis*, albeit less commonly, full-blown septicemia (if the toxins released by the bacteria replicating in the bloodstream are of a sufficient concentration to damage the blood vessels) (sources: Centers for Disease Control and Prevention, Meningitis Research Foundation).

fig. 4 Pictured above are the three classifications for hemolytic properties displayed on a blood agar plate. Alpha-hemolytic bacteria are present in dark-green colonies (due to oxidized hemoglobin), beta-hemolytic bacteria are present in yellow colonies (due to the rupture of red blood cells) and gamma-hemolytic bacteria present in colonies with unchanged colouration. Source: Wikimedia Commons.

More than 90 serotypes of *S. pneumoniae* exist, with 23 serotypes (1, 2, 3, 4, 5, 6B, 7F, 8, 9N, 9V, 10A, 11A, 12F, 14, 15B, 17F, 18C, 19F, 19A, 20, 22F, 23F and 33F) accounting for 88% of strains capable of causing invasive disease in humans. *N. meningitidis* and *S. pneumoniae* together account for 80% of all adult cases of bacterial meningitis (sources: Centers for Disease Control and Prevention, New England Journal of Medicine).

3.1.3 Group B Streptococcus

rupture of red blood cells and gamma-hemolysis indicates the lack of any distinguishable effect. See fig.4

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Group B Streptococcus (abbreviated GBS), also known as *Streptococcus agalactiae*, is a Gram-positive, anaerobic, non-spore forming, beta-hemolytic, spherical bacterium with a group-arrangement varying between diplococcus and streptococcus. The bacteria exist in the gastrointestinal tract of adults, with secondary spreads to other sites, the most important of which is the vagina. GBS can be found in the vaginal flora of 20-30% of all women and may gain access to the amniotic fluid during pregnancy or colonize the newborn as it passes through the birth canal, which occurs in 1-2% of live births (sources: Sherris Medical Microbiology, Meningitis Research Foundation).

GBS infections can take on two forms in newborns: as early-onset within the first six days of life due to the newborn catching the bacteria from the birth canal (as is the case with approximately 60-70% of newborn GBS infections), usually in the form of septicemia with pneumonia (commonly) and/or meningitis (in 5-10% of cases), or as late-onset between one and three months after birth due to other external sources of bacterial contamination, usually in the form of meningitis (sources: Centers for Disease Control and Prevention, Sherris Medical Microbiology, Meningitis Research Foundation).

GBS infections can also occur more uncommonly in adults, as chorioamnionitis[21] and bacteremia in women before or after childbirth or as pneumonia and soft tissue[22] infections in non-pregnant adults (particularly those above 65), male or female, and, rarely in either case, as meningitis. The exact source of infection is unknown. There are 9 serotypes of GBS of which 5 (Ia, Ib, II, III and V) are responsible for the majority of invasive human GBS disease (sources: Centers for Disease Control and Prevention, Sherris Medical Microbiology, Institute Pasteur, PubMed).

3.1.4 Haemophilus influenzae

Haemophilus influenzae (abbreviated *H. influenzae*) is a small, Gram-negative, aerobic, non-spore forming, rod-shaped, bacterium with a coccobacillus group-arrangement that can be found with capsule or without. The bacteria exist in colonies in the normal nasopharyngeal flora of 20 to 80% of the human population. Most of these are nonencapsulated strains (which are less virulent than encapsulated strains), though encapsulated serotypes (a

21 Chorioamnionitis: Bacterial infection and inflammation of the fetal membranes.
22 Soft tissue: All the tissues of the body except bones and organs.

through f) are not rare. Meningitis is the most common form of *H. Influenzae* infection, affecting most commonly children under the age of 2 (source: Sherris Medical Microbiology).

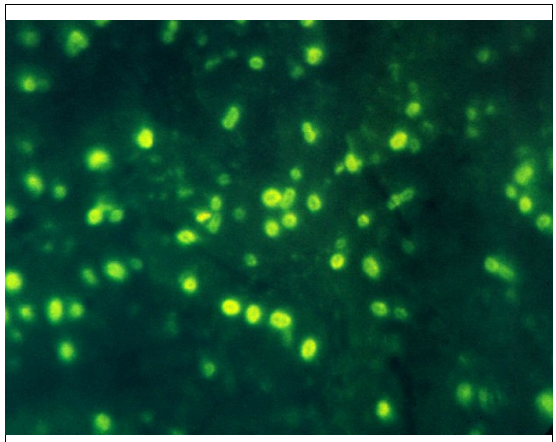

fig. 5 Immunofluorescent micrograph of Haemophilus influenzae. Note the slightly elongated spherical shapes characteristic of the coccobacillus group-arrangement. Source: Centers for Disease Control and Prevention.

The *H. Influenzae* serotype B (abbreviated Hib) is responsible for 90% of cases of *H. influenzae* infections. Using a mechanism that remains to be understood, Hib strains colonising the nasopharyngeal tract occasionally invade into deeper tissues, passing the mucosal barrier. When the bacteria reach the bloodstream (causing bacteremia), they are spread to the central nervous system and other distant sites (such as the bones and joints) via metastatic[23] infections. Meningitis is present in 50% of cases of major acute Hib infections, with the remaining cases distributed among pneumonia, epiglottitis, septicemia and other infections (source: Sherris Medical Microbiology).

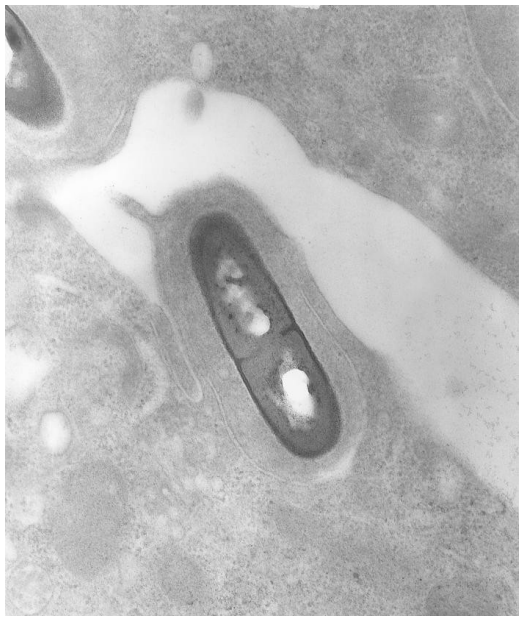

fig. 6 Electron micrograph of Listeria monocytogenes pictured above. Note the rod-shaped bacillus arrangement. Source: Centers for Disease Control and Prevention

As with *N. Meningitidis* and *S. Pneumoniae*, Hib is transmissible via respiratory droplets from the nasopharyngeal tract of carriers of the bacteria or patients through close contact (source: Centers for Disease Control and Prevention).

3.1.5 *Listeria monocytogenes*

Listeria monocytogenes (abbreviated *L. monocytogenes*) is a Gram-positive, aerobic, non-spore forming, rod-shaped, beta-hemolytic, nonencapsulated, bacterium with a bacillus group-distribution. The bacteria is widespread among

23 Metastatic: With transference of disease.

animals in nature (including those associated with the human food supply, such as fowl), and can be found colonising the intestines of 2-12% of the human population. Infection by *L. monocytogenes* is known as Listeriosis, which can be transmitted via foodstuffs (as *L. monocytogenes* can survive and grow at standard refrigerator temperatures), transplacentally to the fetus in pregnancy, or to newborns via the birth canal during childbirth in a manner similar to GBS. The majority of cases occur in infants under a month of age or adults over the age of 60 (source: Sherris Medical Microbiology).

L. Monocytogenes uses a complex method of invading phagocytes and spreading from phagocyte to phagocyte to spread among cells. Listeriosis usually occurs in the form of a stillbirth (if as an intrauterine infection) or a disseminated infection[24] if transmitted near birth or in adulthood. The bacteria have a tropism[25] for the central nervous system, causing meningitis indistinct from other bacterial pathogens (such as *S. pneumoniae* or N. *meningitidis*). There are 11 serotypes of *L. monocytogenes*, with 3 (1/2a, 1/2b, 4b) accounting for the majority of human cases (source: Sherris Medical Microbiology).

3.1.6 *Escherichia coli*

Escherichia coli (abbreviated *E. coli*) is a large, Gram-negative, non-spore forming, rod-shaped, bacterium with encapsulated and nonencapsulated strains, group-arrangements ranging from coccobacilli to elongated bacilli, is a member of the Enterobacteriaceae family, and has a metabolism with no preference for either aerobic or anaerobic conditions (source: Sherris Medical Microbiology).

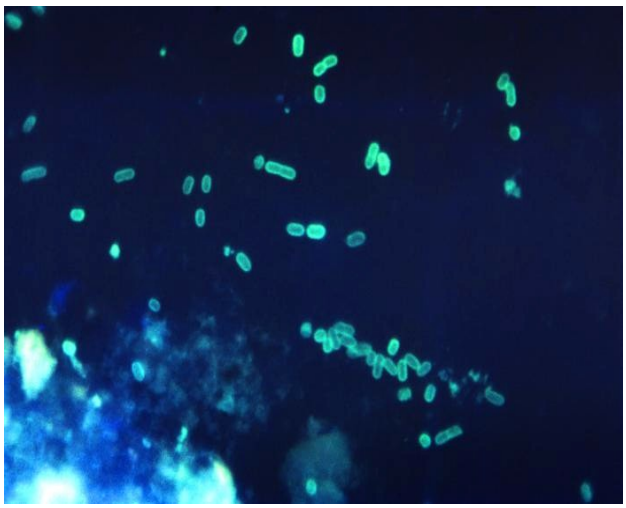

fig. 7 Fluorescent antibody-stain photomicrograph of Escherichia coli. Source: Centers for Disease Control and Prevention.

E. coli can cause a multitude of opportunistic and intestinal infections, having access to every toxin found among the Enterobacteriaceae. Among the opportunistic infections caused by *E.*

24 Disseminated infection: Infection spreading from a single point to throughout the body.
25 Tropism: Preferential target of specific pathogens for their hosts.

coli is neonatal meningitis, causing 20% of all cases and presenting many similar features to GBS disease, in which the bacteria colonise the infant from the vaginal flora (having spread from the patient's own intestinal *E. coli* flora) during childbirth. As with all Gram-negative bacteria, failure to control a local infection of *E. coli* can lead to septicemia and septic shock[26] (sources: Sherris Medical Microbiology, Meningitis Research Foundation).

Serotypes of *E. coli* are derived from three varying antigens: the outer membrane (expressed as O-antigen or simply O), the external capsule (expressed as K-antigen or K), and (in certain strains) flagella (expressed as H-antigen or H). Due to the many distinct O, K, and H antigens, hundreds of serotypes of *E. coli* are possible. Strains possessing the capsule antigen K are responsible for 75% of cases of neonatal meningitis caused by *E. coli* (source: Sherris Medical Microbiology).

3.1.7 *Mycobacterium tuberculosis*

Mycobacterium tuberculosis (abbreviated *M. tuberculosis*) is a Gram-positive (due to the chemical composition of its cell wall, even though M. tuberculosis does not respond to a Gram-stain due to interference from lipids in its cell wall), aerobic, non-spore forming, rod-shaped bacterium with a bacilli group-arrangement (source: Sherris Medical Microbiology).

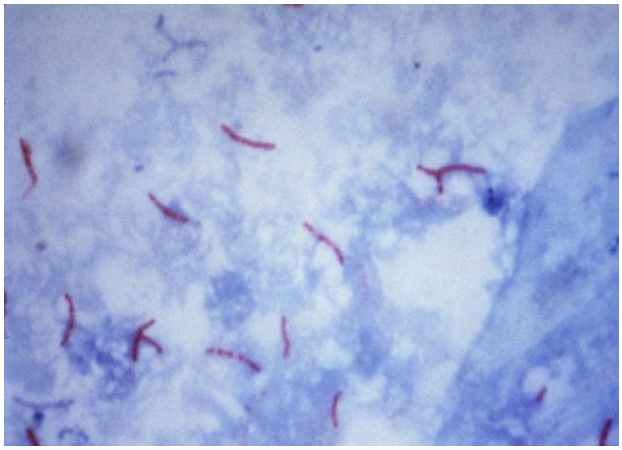

Fig. 8 Ziehl-Neelsen stain photomicrograph of Mycobacterium tuberculosis. Note the thin rod structures and bacilli group-arrangement.
Source: Centers for Disease Control and Prevention.

Humans cannot be carriers of tuberculosis, as the infection needs to develop in the lungs before the patient can transmit the disease. Transmission of the bacteria is due in most part to the inhalation of respiratory droplets carrying the organism, though infection can also occur as a result of ingesting milk from tuberculous cows. In the case of infection through

26 Septic shock: Life-threatening condition involving full-body inflammation due to invasion and persistence of bacteria in the bloodstream. Gram-negative septic shock refers to septic shock caused by endotoxins specific to Gram-negative bacteria.

inhalation of the bacteria, the respiratory droplets are deposited in the respiratory alveoli of the lungs, where they are engulfed by alveolar macrophages[27]. The ingested *M. tuberculosis* bacteria, if the macrophages fail to destroy them, continue replicating, until the macrophages burst and the bacteria are ingested by other blood macrophages. These macrophages can be transported through lymphatic channels, from where they may disseminate to many parts of the body. In infants, the dissemination of *M. tuberculosis* can cause meningitis (sources: Cambridge University Hospitals, Sherris Medical Microbiology).

Tuberculosis progresses to reactivation in 10% of patients recovering from the initial infection at some point in their lives, occurring most commonly in older men. This reactivation can occur in a number of infected sites, including the brain and meninges, which can cause fatal chronic meningitis (source: Sherris Medical Microbiology).

3.2 Prime causative agents of viral meningitis

Viruses are the most prevalent causes of meningitis, with viral meningitis being the disease's most common form. The disease can affect anyone, though children are the most commonly affected. Whilst bacterial meningitis is usually life-threatening, presenting itself alongside septicemia and septic shock, viral meningitis is rarely life-threatening and unusually associated with septicemia. As with bacterial meningitis, there are many types of pathogens capable of causing viral meningitis, of which the major types are detailed here. Due to the general favouring of temperate climates by the most frequent causes of viral meningitis, catching the disease is more common in the summer. Viral meningitis can also be classified as aseptic meningitis, which is meningitis in which bacteria do not grow in cultures of cerebrospinal fluid drawn from the subarachnoid cavity (source: Meningitis Research Foundation).

3.2.1 Enteroviruses

Enteroviruses are a group of very small, unenveloped[28], single-stranded, positive-sense[29],

27 Macrophage: White blood cell subtype of phagocytes.

28 Unenveloped: Viruses can have viral envelopes, modified cell membranes, covering their protein capsids.

29 Positive-sense RNA: Virus RNA strands can be considered positive or negative depending on their ability to be translated by the host cell's ribosomes. Virus RNA can be considered positive-stranded if it can be directly translated, in contrast to negative-stranded RNA, which requires transcription into positive-sense RNA.

Biology

RNA viruses with icosahedral-symmetry capsids. The most common mode of transmission of enteroviruses is via direct or indirect fecal-oral route (contaminated food, contaminated water, insect vectors). Once infection has occurred via this route the virus persists in the oropharynx for between 1 and 4 weeks, and can be shed in the feces for up to 18 weeks. The most common individual method of transmission is person-to-person, suggested by high infection rates in children, with 65% of cases occurring in children under the age of 9. The most frequent incubation period of enteroviruses is relatively short, averaging between 2 and 10 days (source: Sherris Medical Microbiology).

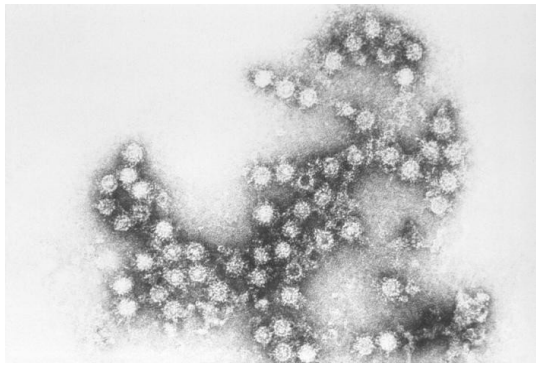

fig 9. Immunoelectron micrograph of enterovirus Coxsackie B4. Source: Centers for Disease Control and Prevention.

Enteroviruses follow a lytic cycle of reproduction: they bind to the surface of a cell and, following adhesion, enter the cell via endocytosis, begin protein synthesis at the ribosomes, and induce cell lysis, releasing the newly-produced virions. After this initial reproductive cycle, which takes place in epithelial cells and lymphoid tissues of the upper respiratory and gastrointestinal tracts, the infection spreads through viremia[30] to other sites, varying by strain, such as the central nervous system, heart, liver, lungs, or pancreas (source: Sherris Medical Microbiology).

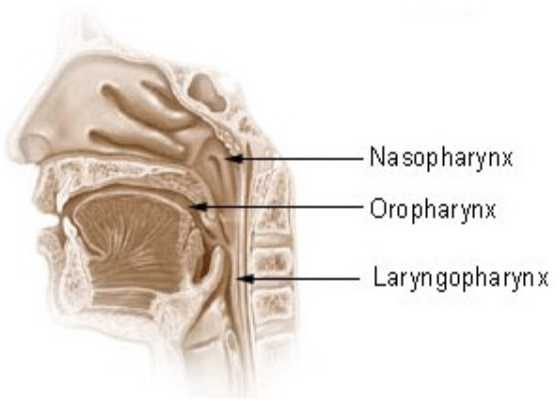

fig. 10 Depicted above is the human pharynx, note the position of the nasopharynx behind the nose and oropharynx behind the uvula. Source: Wikimedia Commons.

Half of all cases of aseptic meningitis are caused by two members of the enterovirus family, both of which have an increased tendency to affect the meninges: coxsackieviruses (which can be classified into Group A and Group B) and echoviruses. Of the 29 serotypes of coxsackievirus A and B, 5 serotypes from group A (2, 4, 7, 9, 10) and 5 serotypes from group B (1 through 5) can lead to aseptic meningitis, and of the 28

30 Viremia: Presence of pathogenic viruses in the bloodstream.

serotypes of echovirus, 6 (4, 6, 9, 11, 16, 30) can lead to aseptic meningitis. Approximately 60% of infections caused by these two viruses are subclinical, presenting only minor symptoms. Aseptic meningitis is the most common clinical disease associated with enteroviruses, usually as a mild condition lasting between 5 and 14 days (source: Sherris Medical Microbiology).

3.2.2 Mumps viruses

Mumps viruses are single-stranded, enveloped, negative-sense RNA viruses with a helical-symmetry capsids. Infection from mumps viruses is observed with most frequency between ages 5 and 15, and can occur in any season, though most frequently in late winter and spring. The virus replicates locally after entering into the respiratory tract, which is followed by dissemination of the virus, through viremia, to target tissues such as the salivary glands and the central nervous system (source: Sherris Medical Microbiology).

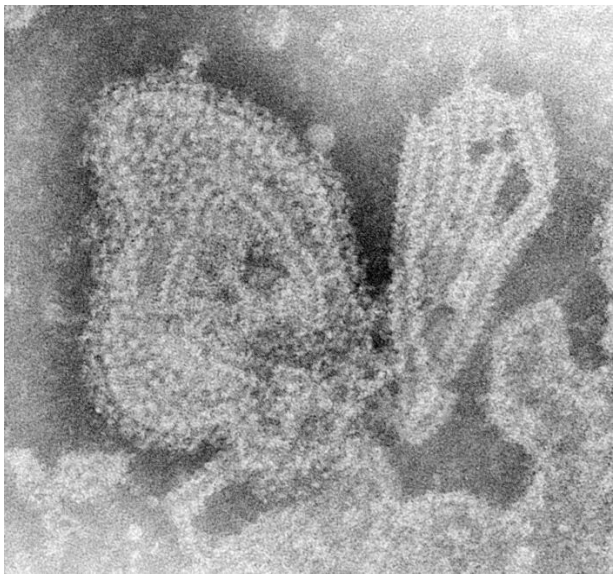

fig. 11 Transmission electron micrograph of mumps virus. Source: Centers for Disease Control and Prevention.

The disease is highly infectious, being acquired in 85% of susceptible contacts, and can be spread from person-to-person via aerosol from the infected respiratory system approximately from 7 days before the onset of symptoms until 9 days after, though the disease only develops from the infection in 60% to 70% of contacts. The average incubation period of mumps viruses before the onset of symptoms is 16 to 18 days. Viruria, the presence of viruses in the urine, is common with mumps disease, and can be detected for up to 14 days following the onset of the illness. Mild, aseptic meningitis is a complication of mumps disease that occurs in 10% of cases between 1 and 3 weeks after the onset of symptoms, though permanent effects can occur as a result of severe infection of the central nervous system (Source: Sherris Medical Microbiology).

3.2.3 Herpesviruses

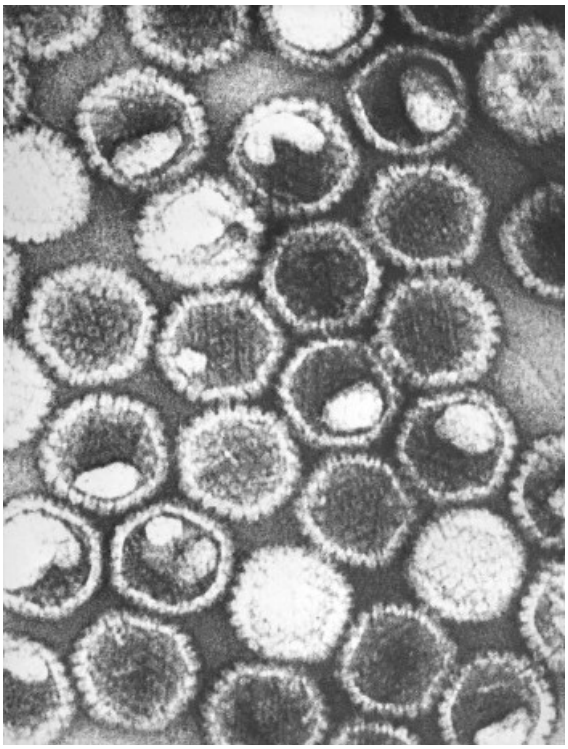

fig. 12 Transmission electron micrograph of herpes simplex virions. Source: Centers for Disease Control and Prevention.

Herpesviruses are a group of large, enveloped, double-stranded DNA viruses with icosahedral-symmetry capsids. Herpesviruses can be classified into three groups: alpha (Herpes simplex viruses 1 and 2 and Varicella-zoster virus, abbreviated HSV-1, HSV-2 and VZV, respectively), beta (Cytomegalovirus and Human herpes virus 6-7, abbreviated CMV, HHV-6 and HHV-7, respectively), and gamma (Epstein-Barr virus and Human herpesvirus-8, abbreviated EBV and HHV-8, respectively). In its replication cycle, the virus enters a cell by fusion of its capsid with the host's cell membrane, which is followed by the migration of the virus' DNA to the nucleus, where it is transcribed to messenger-RNA. The virions synthesised with the translated proteins are released via exocytosis. In all herpesviruses except CMV, the host cell's metabolism is shut down by the virus, leading to cell death (source: Sherris Medical Microbiology).

Herpes simplex viruses appear only in humans, and are transmitted via direct contact with infected secretions. HSV-1 and HSV-2 are causative agents of genital herpes, a sexually-transmitted disease. HSV-2 is more often associated with genital herpes, as it causes 70% of all cases, whilst HSV-1 is more often associated with oral disease. Of the 10% of all cases of clinically evident genital HSV, 1% develop aseptic meningitis. EBV can be found in the saliva of between 10 and 20% of healthy adults. EBV infection is known as infectious mononucleosis, and is transmitted through repeated close-contact. Initially, EBV infects epithelial cells, followed by B lymphocytes. Complication of infectious mononucleosis can occur in 1 to 5% of cases of clinical infection, including encephalitis and aseptic meningitis (source: Sherris Medical Microbiology).

3.2.4 Arboviruses

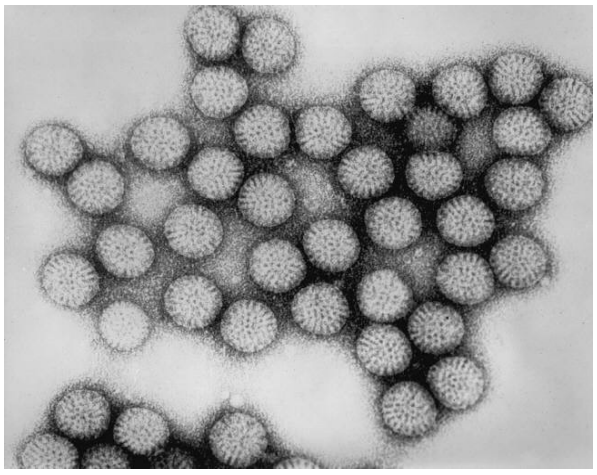

fig. 13 Transmission electron micrograph of rotavirus virions, a genus pertaining to the Reoviridae family. *Source: Centers for Disease Control and Prevention.*

Arboviruses are viruses transmitted via arthropod vectors, through infected bloodsucking insects such as mosquitoes or ticks. Arboviruses cover a wide range of families, such as togaviruses and flaviviruses (enveloped, icosahedral-symmetry single-stranded, positive-sense RNA viruses), bunyaviruses (enveloped, helical-symmetry, single-stranded, negative-sense RNA viruses), reoviruses (unenveloped, helical-symmetry, double-stranded RNA viruses), arenaviruses (enveloped, complex-symmetry, single-stranded, negative-sense RNA viruses) and filoviruses (enveloped, helical-symmetry single-stranded, negative-sense RNA viruses) (source: Sherris Medical Microbiology).

In the majority of transmissions, the arbovirus is not sustained solely in the vector species, instead being found and sustained through viremia in nonhuman vertebrate reservoirs from which the vector can reacquire the virus and then transmit to human hosts. Clinical manifestations of arbovirus infections are classified into three major groups: hemorrhagic fever (damage to small blood vessels), affecting major organ systems, and affecting the central nervous system, which has as a consequence aseptic meningitis or meningoencephalitis[31]. Infection of the central nervous system occurs as a result of viremia following a bite by an infected arthropod, with the virus crossing the blood-brain barrier (source: Sherris Medical Microbiology).

3.3 Prime causative agents of fungal meningitis

Fungal meningitis is the inflammation of the meninges caused by a fungal pathogen. It is not usually contagious from person-to-person and is acquired in most cases from environmental spores (source: Centers for Disease Control and Prevention).

31 Meningoencephalitis: Inflammation of the meninges and, simultaneously, inflammation of the brain.

3.3.1 *Cryptococcus neoformans*

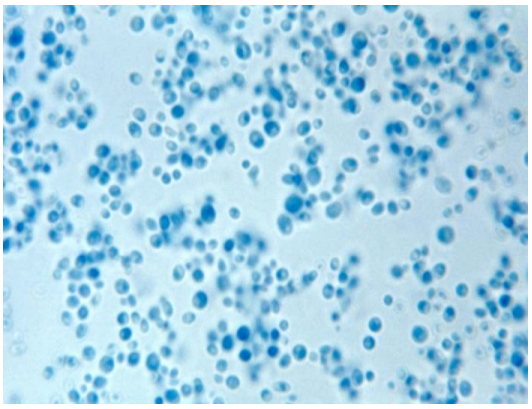

fig. 14 Photomicrograph of the yeast form of Cryptococcus neoformans. Source: Centers for Disease Control and Prevention.

Cryptococcus neoformans (abbreviated *C. neoformans*), also known as cryptococcus, is an infectious capsuled yeast[32]. Whilst not an opportunistic fungi[33], infection from *C. neoformans*, cryptococcosis, also known as cryptococcal disease, occurs most seriously in immunocompromised patients. *C. neoformans* is present in soil contaminated from bird feces all around the world, with infection in humans presumed to occur as a result of the inhalation of the aerosolised cryptococci. Once inhaled, the yeast resist macrophages in the lung by overproducing a polysaccharide external capsule. In the lung, *C. neoformans* causes pneumonia as its primary disease, and, once disseminated, cryptococcus has an affinity for the central nervous system, where it causes chronic meningitis, which accounts for the majority of cases of fungal meningitis. The incubation time for *C. neoformans* is unknown, though the average onset of symptoms of cryptococcosis is estimated to occur between 2 and 11 months after exposure to the yeast cells (sources: Meningitis Research Foundation, Centers for Disease Control and Prevention, Sherris Medical Microbiology).

3.3.2 *Candida albicans*

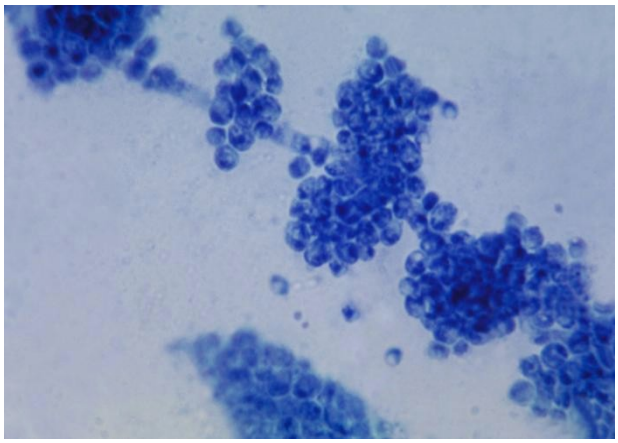

fig. 15 Photomicrograph of Candida albicans. Source: Centers for Disease Control and Prevention.

Candida albicans (abbreviated *C. albicans*) is an infectious yeast. Infection from *C. albicans*, candidiasis, is opportunistic can occur in local and disseminated forms as a result of the shift in structure of the fungi from yeast to pathogenic, though the precise trigger of this shift in humans is

32 Yeast: Single-celled form of fungus.

33 Opportunistic fungi: Pathogenic fungi normally incapable of causing infection and disease to healthy people.

unknown. *C. albicans* can be commonly found in floras such as the oropharyngeal and gastrointestinal flora. In very rare cases of the infection, fungal meningitis can occur through dissemination of the fungi through the blood to the spinal cord (source: Sherris Medical Microbiology, Centers for Disease Control and Prevention).

3.4 Prime causative agents of non-infectious meningitis

An inflammation of the meninges that does not stem from an infective agent is classified as non-infectious meningitis. It cannot be spread by any form of person-to-person contact (source: Centers for Disease Control and Prevention).

3.4.1 Neoplastic meningitis

Neoplastic meningitis is the development of meningitis due to the infiltration of the subarachnoid space with cancer cells. Neoplastic meningitis occurs in approximately 5% of all cancer patients, and is the third most common site within the central nervous system for cancer to metastasise to (source: Neurologist, PubMed).

3.4.2 Drug-induced aseptic meningitis

Drug-induced aseptic meningitis (abbreviated DIAM) is the development of meningitis due to the administration of certain medications. Four groups of medications provide the most frequent causes of DIAM: nonsteroidal anti-inflammatory drugs (abbreviated NSAIDs), antibiotics, intravenous immunoglobulins and OKT3 antibodies[34]. The precise mechanism that causes the inflammation of the meninges is not understood, though is generally believed to be an immunologic hypersensitivity reaction. Ibuprofen is the most frequently cited drug as a cause of DIAM among the NSAIDs (source: Archives of Internal Medicine, PubMed).

3.4.3 Systemic lupus erythematosus

Systemic lupus erythematosus (abbreviated SLE) is a chronic autoimmune disorder. The

34 OKT3 antibody: Mouse monoclonal (a type of homogenous antibodies) antibody used to treat transplant rejection.

underlying cause of SLE (and all autoimmune diseases) is not fully understood, though SLE may be caused by certain drugs (in such an occurrence, it is classified as drug-induced lupus erythematosus). The disease affects women nine times as often as men and people of African and Asian descent are affected more often. SLE can cause vasculitis[35], affecting the skin, joints and organs, and, in rare cases, non-infectious meningitis (sources: Centers for Disease Control and Prevention, PubMed).

4. Features and general symptoms of meningitis

4.1 Features and general symptoms of bacterial meningitis

The characteristic symptoms of bacterial meningitis are the onset (sudden or gradually over several days) of fever, headache, and neck stiffness, often alongside other symptoms such as nausea, vomiting, photophobia[36] or an altered mental status. These symptoms typically develop within 3 and 7 days after exposure (source: Centers for Disease Control and Prevention).

4.1.1 Features and general symptoms of bacterial meningitis in adults

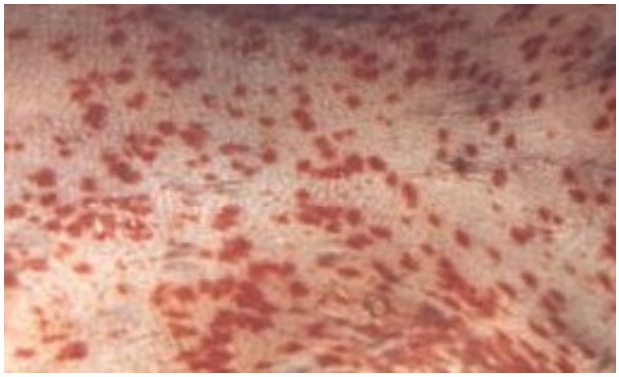

fig. 1 Image adapted from a photograph depicting the shaved anterior thoracoabdominal region, displaying a petechial rash, of a rock squirrel afflicted with the plague. Source: Centers for Disease Control and Prevention

In a study published in the New England Journal of Medicine in 2004 that defined adults as patients older than 16 years, 696 episodes of community-acquired[37] adult bacterial meningitis in 671 adults were drawn from the Netherlands Reference Laboratory for Bacterial Meningitis from between October 1998 and April 2002. Relating to symptoms on presentation, 83% of episodes (569 out of 685 evaluated) presented **neck stiffness**, 77% (522/678) presented **fever** (defined here as a

35 Vasculitis: Inflammation and destruction of the blood vessels.
36 Photophobia: Abnormal sensitivity to light.
37 Community-acquired meningitis: Meningitis acquired in the community, in contrast to nosocomial meningitis, meningitis acquired in hospital.

body temperature equal to or superior to 38ºC), 69% presented a **change in mental status**, defined by a score below 14 on the Glasgow Coma Scale[38] (this symptom alongside the aforementioned two making up the classic triad of symptoms, something which only 44% of episodes accounted for), 96 of these presenting on admission (14% overall) a score below 8, indicating that they were comatose, and 87% (544/626) presented **headaches**. Two of the four symptoms listed above were present in 95% of episodes, a single symptom was present in 4% of episodes, and none of the four symptoms were present in 1% of episodes. Patients were comatose on admission in 14% of episodes. **Focal neurological deficits**[39] were present in 33% of episodes and others developed during the clinical course, encompassing 351 (50%) of episodes. Seizures occurred in 5% (32/666) of episodes before admission and in 15% (107/696) during the clinical course (source: New England Journal of Medicine, PubMed).

Of the 696 analysed episodes of community-acquired meningitis, 352 (51%) were due to *S. pneumoniae* (with 35 different serotypes identified, 3 and 14 being the two most prominent, being responsible for 36 and 34 episodes, respectively), 257 (37%) were due to *N. meningitidis* (with group B identified in 173 episodes, C in 79, Y in 3, H in 1, and W135 in 1), 30 (4%) were due to *L. monocytogenes*, and 57 (8%) were due to other bacteria [*H. influenzae* (14), *Staphylococcus aureus* (9), Group A Streptococcus (6), GBS (5), *Streptococcus suis* (4) and *E. coli* (4) causing 74% of these] (source: New England Journal of Medicine, PubMed).

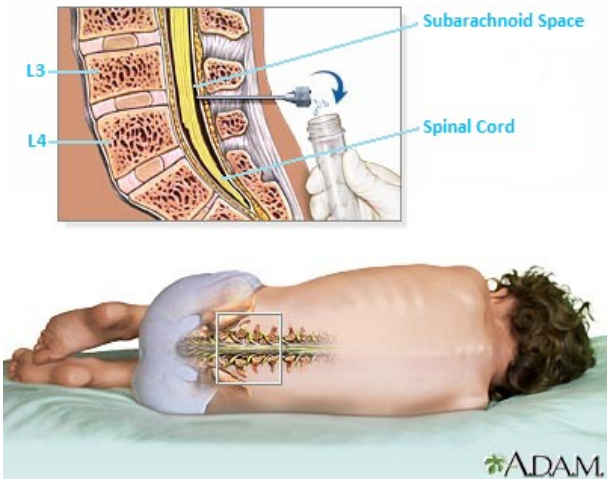

fig. 2 Image depicting the lumbar puncture procedure. Source: Adapted from an A.D.A.M.,Inc. image.

The classic triad was more than twice as frequent in patients with pneumococcal meningitis than those with meningococcal meningitis, appearing in

38 Glasgow Coma Scale: Neurological scale encompassing motor elements, verbal elements, and the ability to open one's eyes, in which a patient is assigned a score between 3 (deep unconsciousness, comatose) and 15 (minor or no injury).

39 Focal neurological deficit: Impairments in behaviour or perception caused by lesions in particular (focal) areas of the central nervous system, such as loss of memory, balance, loss of auditory, visual or tactile sensation, or an impaired motor function.

58% and 27% of episodes, respectively, with P<0.001 (a statistical certainty of 99.9%). Additional symptoms include nausea, present in 74% (449/610) of episodes, and a **rash**, present in 26% (176/683) of episodes, with *N. meningitidis* being the causative species in 162 of the 176 episodes of rash, of which 144 were petichial[40]. The causative species in the remaining 14 episodes of rash (13 of which were petechial) was *S. pneumoniae* in 9, GBS in 3, and *H. influenzae* and *L. monocytogenes* in 1 each (source: New England Journal of Medicine, PubMed).

Lumbar puncture[41] was performed in all patients, and in the 645 episodes in which cerebrospinal fluid drawn was analysed, 7% (47) of episodes presented a white blood cell count below 100/mm^3, 14% (93) presented one between 100 and 999/mm^3 and 78% presented one above 999/mm^3 (505), with the mean **white blood cell count** at 7753/mm^3, with a standard deviation (abbreviated SD; with 68.2% of all cases occurring within a range) of ±14736. The average **protein level** analysed was 4.9g/L, with a SD of ±4.5, and the average cerebrospinal fluid **glucose to blood glucose ratio** was 0.2, with a SD of ±0.2. There was at least one of four individual cerebrospinal fluid values (a cerebrospinal fluid glucose to blood glucose ratio below 0.23, a glucose level below 0.34g/L, a protein level above 2.2g/L, or a white blood cell count above 2000/mm^3) predictive (capable of ruling in with 99% certainty, established via a study of 422 patients with bacterial or viral meningitis done by Duke university and published in the Journal of American Medical Association in 1989) of bacterial meningitis in 88% (567) of the 645 episodes analysed. In the 611 patients in which a blood culture was performed, 66% (404) presented a positive blood culture. The average blood serum **C-reactive protein[42] concentration**, which was determined in 394 patients, was 225mg/L, with a SD of ±132. Additionally, in the 386 episodes that underwent an opthalmoscopy, 13 (3%), were characterised by papilledema[43] (source: New England Journal of Medicine, PubMed).

40 Petichial rash: Rash composed of small purplish spots caused by minor hemorrhages, caused by the breaking of capillary blood vessels. See fig. 1

41 Lumbar puncture: Diagnostic procedure performed to collect a sample of cerebrospinal fluid between two of the lumbar vertebrae, usually L3 and L4, that is then analysed. See fig. 2 for procedure and fig. 3 for normal cerebrospinal fluid values in comparison with values yielded when the patient is afflicted by various types of meningitis.

42 C-reactive protein: Protein synthesised by the liver and secreted into the bloodstream within 6 hours after an acute inflammatory reaction.

43 Papilledema: Edema of the optic disk, potentially caused by increased intracranial pressure.

Condition	Glucose	Protein	Cells
Normal Values	50-66% of blood glucose	0.15-0.45g/L	0-4/mm^3 WBC – 0/ mm^3 RBC
Bacterial Meningitis	Low | Elevated lactate	High	>300/mm^3 PMNs
Viral Meningitis	Normal	Normal or High	<300/mm^3 mononuclear
Tuberculous Meningitis	Low	High	<300/mm^3 PMNs–mononuclear
Fungal Meningitis	Low	High	<300/mm^3 mononuclear
Malignant Meningitis	Low	High	mononuclear

fig. 3 Table depicting the comparison between cerebrospinal fluid analyses in patients with various types of meningitis. WBC: White blood cell – RBC: Red blood cell – PMNs: Polymorphonuclear leukocytes, the most abundant type of WBC in mammals – Mononuclear: WBC with a one-lobed nucleus, comprising monocytes and lymphocytes. Source: Oxford Handbook of Clinical and Laboratory Investigation.

At the time of discharge, all patients were graded according to the Glasgow Outcome Scale[44] and 550 of the 553 surviving patients received a neurological examination. Of the 696 episodes of community-acquired bacterial meningitis, 21% (143) resulted in death, which included 30% (107/352) of pneumococcal meningitis episodes, 7% (19/257) of meningococcal meningitis episodes and 20% (17/87) of meningitis episodes with other causative agents. Additionally, 66% (459/696) of episodes were discharged with mild or no disabilities, which included 50% (175/352) of pneumococcal meningitis episodes, 88% (227/257) of meningococcal meningitis episodes and 66% (57/87) of meningitis episodes with other causative agents. The most common disabilities reported were hearing loss in 14% (78/550) of episodes and hemiparesis[45] in 4% (24) (source: New England Journal of Medicine, PubMed).

In a similar study published in the New England Journal of Medicine in 1993 which examined the clinical features of 259 adults diagnosed with bacterial meningitis between January 1962 and December 1988, the results were similar, with the diagnostic triad of fever, neck stiffness and a change in mental status being just as if not more frequent in examined patients. As with the first study, *S. pneumoniae* was the leading cause followed by *N. meningitidis*. Additionally, cerebrospinal fluid analyses showed similar results, matching the values

44 Glasgow Outcome Scale: 5-point scale used to generalize the recovery of a victim of brain trauma, in which a score of 1 indicates death, 2 indicates a vegetative state, 3 indicates severe disability (consciousness without independent living), 4 indicates moderate disability (independent living), 5 indicates mild or no disability.

45 Hemiparesis: Mild paralysis affecting one side of the body.

depicted in figure 3. The analysis of this second study serves to support the results garnered in the first study (source: New England Journal of Medicine, PubMed).

4.1.2 Features and general symptoms of bacterial meningitis in children

In a study published in the Tropical Medicine & International Health journal in 1998, various clinical features of 267 children, defined as patients below the age of 14, were examined at the Queen Elizabeth Central Hospital in Malawi during a one-year period between 1996 and 1997. There were 61 patients in the neonatal group, defined as patients below the age of one month, and 206 in the post-neonatal group, which were further split into age groups of 1-12 months (102 patients), 1-5 years (59 patients), and 5-14 years (45 patients). **Fever** was present in the histories of 87% of neonates and 89, 91, and 95% of post-neonates, **neck stiffness** was present in 12% of neonates and 44, 63, and 86% of post-neonates, **headache** was not applicable to neonates and present in 6, 25, and 78% of post-neonates. **Altered consciousness**, defined in this study as a score below 5 on the Blantyre Coma Scale[46], was present in 58% of neonates and 55, 51, and 71% of post-neonates, seizures were present in 25% of neonates and 42, 47, and 46% of post-neonates, and **vomiting** was present in 15% of neonates and 34, 44, and 33% of post-neonates. **Bulging fontanelle**[47] was present in 67% of neonates and 49 and 16% of post-neonates, being inapplicable in the 5-14 years age group (source: Tropical Medicine & International Health, PubMed).

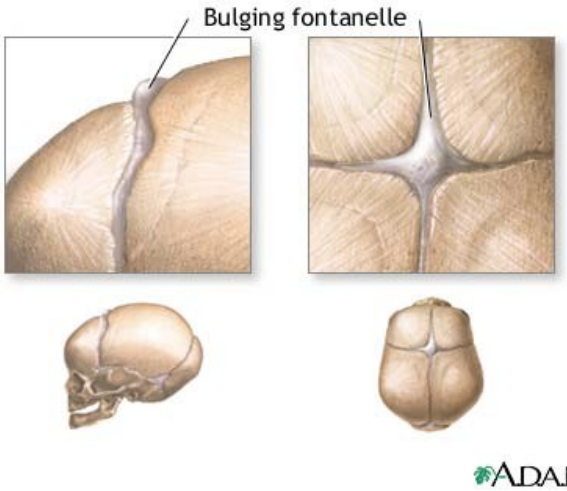

fig. 4 Image depicting the outward curving of an infant's fontanelle caused by elevated intracranial pressure and associated with, amongst other things, meningitis. Source: A.D.A.M. Inc.

The overall mortality rate was 40%, with *S. pneumoniae* and *H. influenzae* being the most common causative agents of bacterial meningitis [detected in 23%

46 Blantyre Coma Scale: Neurological scale encompassing motor elements, verbal elements, and eye movement in which a patient is assigned a score between 0 (unresponsive) and 5 (normal results).

47 Bulging fontanelle: Bulging of the soft membranous spot on the head of a neonate or infant, indicative of increased intracranial pressure. See fig. 4

(62/267) of episodes and 17% (44/267), respectively] and simultaneously presenting the highest mortality rates [with 46% (27/59 with recorded outcomes) and 43% (18/42), respectively] (source: Tropical Medicine & International Health, PubMed).

There are multiple key differences in setting between the three studies analysed. In the first, only episodes in which cerebrospinal fluid cultures were positive were used for the study, whereas in the other two a compatible clinical picture and a pleocytosis[48] meeting a certain criteria was sufficient. In the second, approximately one quarter of community-acquired episodes were culture-negative and in the third, 29% of episodes were included. In patients with bacterial meningitis, negative cerebrospinal cultures occur in between 11 and 30% of episodes, though there have been no reported significant clinical differences between culture-negative and culture-positive bacterial meningitis. The setting of the third study was implicated in the elevated mortality rates, with the presence of *H. influenzae* (reduced by up to 98% in many parts of the world, including the setting of the first study) and malnutrition being prominent factors, and potentially the lack of rash symptomatology recording, as N. meningitidis is uncommon in southern Malawi and therefore was rarely seen in the study (sources: New England Journal of Medicine, Tropical Medicine & International Health, PubMed).

Kernig's sign[49] and Brudzinski's sign[50] are two diagnostic signs commonly associated with meningitis, but have not been fully investigated. In a study published in the Clinical Infectious Diseases journal in July 2002, the diagnostic accuracy of Kernig's sign and Brudzinski's sign in discriminating between patients with and without meningitis was evaluated in 297 adults (defined as patients older than 16 years) who presented to the Yale-New Haven Hospital Emergency department between July 1995 and June 1999 with suspected meningitis (defined as the presence of symptoms corresponding with meningitis such that a lumbar puncture was performed). Approximately 5% (3/66) of patients with confirmed meningitis presented Kernig's sign, as did 5% (8/163) of those who did not have meningitis, and the same was true for Brudzinski's sign (5% and 5% respectively) (source: Clinical Infectious Diseases, PubMed).

48 Pleocytosis: Increased presence of cells, in this case, in cerebrospinal fluid.

49 Kernig's sign: Sign devised to indicate meningeal irritation, found positive when the leg is bent at the hip and knee at 90º angles and subsequent extension of the knee results in pain. See fig. 5

50 Brudzinski's sign: Sign devised to indicate meningeal irritation, found positive when the passive flexion of the neck results in flexion of the hips and knees. See fig. 5

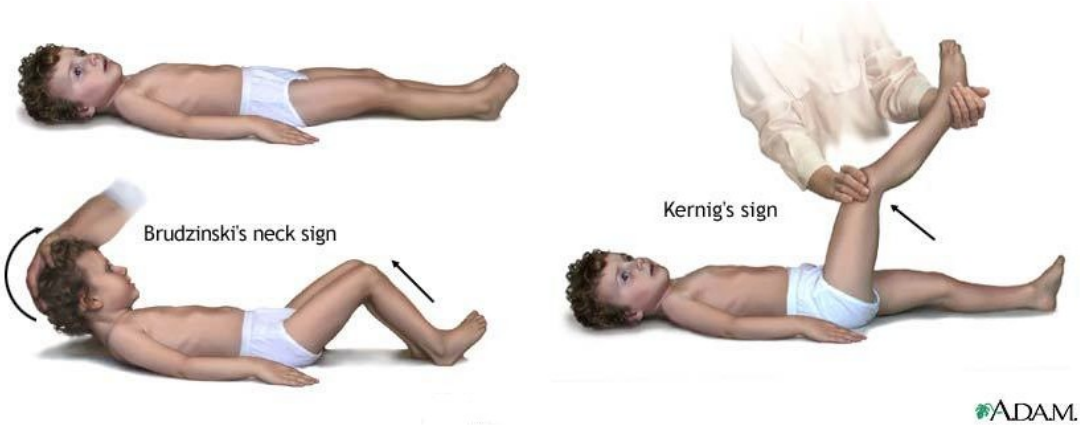

fig 5. Image depicting Brudzinski's sign (left) and Kernig's sign (right), diagnostic signs commonly associated with meningism, or meningeal irritation. *Source: Adapted from A.D.A.M.,Inc. images.*

4.2 Features and general symptoms of aseptic and viral meningitis

The characteristic symptoms of viral meningitis are similar to those of bacterial meningitis but present a varied range of appearance rates depending on the causative agent.

4.2.1 Features and general symptoms of aseptic and viral meningitis in children

In a study published in the PLoS ONE scientific journal in 2007 that defined children as patients younger than 14 years, the clinical features 506 children diagnosed with viral or aseptic meningitis between 1994 and 2002 at the Aghia Sophia Children's Hospital in Athens were evaluated. Relating to symptoms on admission, 98% of patients presented **fever**, 94% presented **headache**, 67% presented **vomiting**, 60% presented **neck stiffness**, 46% presented lethargy or irritability, 40% presented anorexia, and 9% presented **rash**. In all 506 cases there were no reported serious complications or deaths (source: PLoS ONE, PubMed).

Cerebrospinal fluid analyses were conducted in all patients of the study. Relating to **white blood cell count**, 4% (19) of patients presented one between 10 and 25/mm^3, 18% (91) presented one between 26 and 100/mm^3, 57% (289) presented one between 101 and 500/mm^3, 15% (75) presented one between 501 and 1000/mm^3, and 6% (32) presented one above 1000/mm^3. The medium (50th percentile) white blood cell count was 201, with an interquartile (between the 25th and 75th percentile) range of 117-417. The medium **protein**

level analysed was 0.34g/L, with an interquartile range of 21-53, and the medium **glucose level** analysed was 0.53g/L, with an interquartile range of 47-60. There were no observed differences in cerebrospinal fluid compositions between agegroups excepting children younger than twelve months, who presented higher protein levels and lower glucose levels. The cerebrospinal fluid of 96 children was analysed through PCR[51] and 47 samples tested positive for enterovirus RNA (source: PLoS ONE, PubMed).

4.2.2 Features and general symptoms of aseptic and viral meningitis in adults

In a study published in the Critical Care medical journal in 2011, the clinical features of 218 patients diagnosed with viral meningitis were recorded at the emergency unit of the Saint-Etienne University Hospital between 1997 and 2009. The mean age of the patients admitted to the study was 35 with a SD of ±18. Relating to symptoms of presentation, the mean **body temperature** recorded was 39.1ºC with a SD of ±0.2, **headache** was present in 72% (158) of patients, **neck stiffness** was present in 55% (121), **confusion** was present in 14% (31), and the mean Glasgow Coma Scale score was 14 with a SD of ±2. Additionally, the average **C-reactive protein concentration** was determined to be above 30mg/L in 25% of episodes of viral meningitis, with an average concentration of 42mg/L and a SD of ±39 with a range of 3-152 (source: Critical Care, PubMed).

4.3 Features and general symptoms of fungal meningitis

Recurring fungal meningitis is the primary disease caused by cryptococcosis (disease caused by *C. neoformans*) and has similar clinical presentations to candidiasis (disease caused by *C. albicans*) and tuberculous meningitis. Most cases of fungal meningitis occur in patients presenting immunodeficiency. The common clinical manifestation of this type of recurring meningitis is increasingly severe headaches over several weeks with other symptoms common to the majority of meningitis episodes, such as neck stiffness, behavioural changes, photophobia, nausea and, less commonly, seizures, hearing loss and muscle pain. Fever is present in roughly half of such episodes of meningitis, though is more frequent in patients

51 PCR: Polymerase Chain Reaction, a technique for amplifying one or several pieces of DNA or RNA that serves multiple purposes, including detection for the presence of infectious diseases.

with acquired autoimmune deficiency syndrome (which is also usually accompanied by a more rapid onset of symptoms) (sources: Cecile Medicine, Sherris Medical Microbiology, Meningitis Research Foundation).

5. Diagnosis, treatment and prevention of meningitis

5.1 Diagnostic techniques for the detection of meningitis

5.1.1 Diagnostic techniques for the detection of bacterial meningitis

In patients presenting symptoms indicative of bacterial meningitis, the suggested course of action is to obtain blood samples for culture and to perform a lumbar puncture immediately to evaluate whether the cerebrospinal fluid extracted is consistent with bacterial meningitis. When a lumbar puncture is delayed or a head CT scan[52] is required, blood samples should be obtained and the appropriate empirical antimicrobial treatment[53] and adjunctive therapy[54] due to the increased danger acute bacterial meningitis poses if left untreated for too long (source: Clinical Infectious Diseases, PubMed).

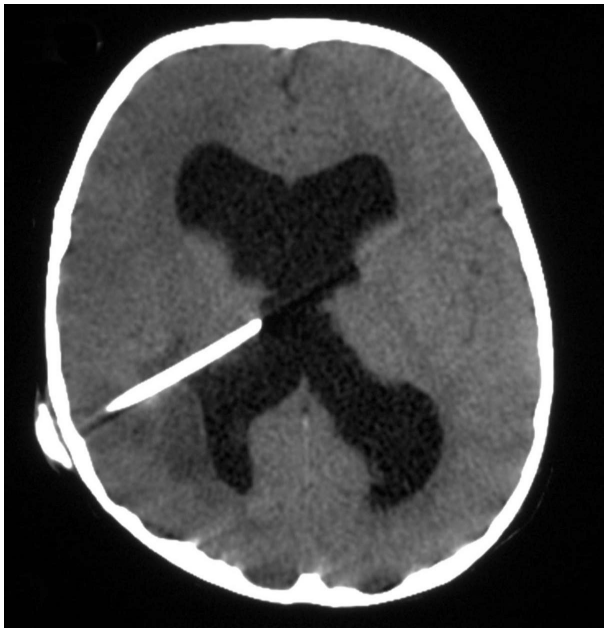

fig. 1 Head CT scan showing an abscess, which acts as a mass, increasing intracranial pressure, with a cerebral shunt inserted, draining an excess build-up of cerebrospinal fluid. Source: Wikimedia Commons.

A lumbar puncture can lead to brain herniation in patients with elevated intracranial pressure, which is precipitated especially in patients with intracranial mass lesions[55] (though the rate of this complication is unknown). A head CT scan is issued when clinical features indicate an intracranial mass lesion or another cause of elevated intracranial pressure in order to evaluate the risk of brain herniation as a result of a lumbar puncture. There are

52 CT scan: X-ray computed tomography scan, a method of medical imaging. See fig. 1

53 Empirical treatment: Treatment issued before a firm diagnosis is established.

54 Adjunctive therapy: Secondary therapy issued alongside the primary treatment with the objective of increasing the primary treatment's effectiveness.

several criteria which may be taken to justify a head CT scan, including an immunocompromised state, a history of CNS diseases, any recent seizures, neurosurgery, papilledema, an abnormal level of consciousness, or a focal neurological deficit (source: New England Journal of Medicine, Clinical Infectious Diseases, PubMed).

Whilst the results of cultures derived from cerebrospinal fluid of patients with bacterial meningitis who have not received antimicrobial treatment are positive in between 70 and 85%, cultures take between 24 and 48 hours to isolate the causative organism. As a result of this, several, more rapid diagnostic tests can be considered to begin with more specific antimicrobial treatment sooner, or to eventually discard bacterial meningitis as a diagnosis and end antimicrobial treatment. A **Gram stain** of the cerebrospinal fluid can identify the causative organism accurately in between 60 and 90% of patients with community-acquired meningitis, and in approximately 20% fewer of patients who experienced antimicrobial treatment prior to the lumbar puncture. Though the utility of a Gram stain varies depending on the causative organism, Gram stain evaluation is a recommended evaluation to perform in all patients with suspected meningitis. If the Gram stain is negative, empirical antimicrobial treatment is continued, unless viral meningitis has been confirmed (Sources: Infectious Disease Clinics of North America, Clinical Infectious Disease Journal, PubMed).

Other rapid diagnostic tests include **latex agglutination**[56], which presents a good sensitivity towards the antigens of common causative organisms of bacterial meningitis (which varies, as in the case of Gram staining, depending on the bacteria), though discouraged by the Practice Guideline Committee due to a lack of treatment modification in the majority of cases where the latex agglutination test was positive, and a number of uncommon cases

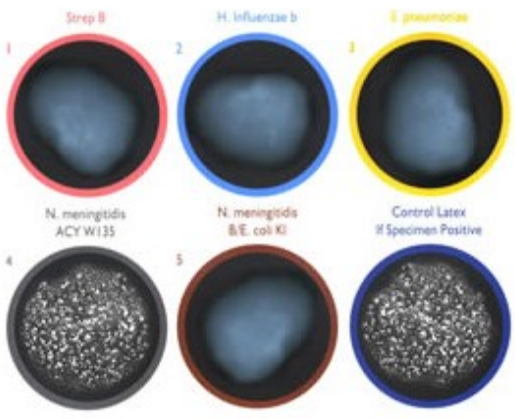

fig. 2 Series of bacterial agglutination assays, with a positive result for N. meningitidis of serotype A, C, Y, or W135. Note the final assay is a control used for further confirmation.
Source: Oxoid

55 Intracranial mass lesion: Space-occupying lesion in the head, which increases intracranial pressure. See fig. 1
56 Latex agglutination: Diagnostic procedure in which tiny particles of latex coated with a specific antibody

in which false-positive test results led to unnecessary additional treatment as well as prolonged hospitalisation. The test is recommended by some and is shown to be most useful in cases with negative Gram stain and CSF culture results and where the patient has already received empirical antimicrobial treatment (source: Clinical Infectious Diseases, PubMed).

Cerebrospinal fluid **PCR**, when used as a diagnostic tool for identifying the common bacterial causative agents of meningitis, presents high sensitivity and specificity. PCR utilising a broad range of bacterial primers has shown a very high negative predictive values[57]; in a study published in 2003 based on 74 cerebrospinal fluid samples from 70 patients, broad range PCR (using a primer derived from the bacterial 16S ribosomal DNA gene) presented a negative predictive value of 100%. Because of this, broad-range bacterial PCR can be used to exclude bacterial meningitis as a possible diagnosis and influence decisions to discontinue antimicrobial treatment. The use of PCR to discover the causative organism of meningitis is recommended in the event of a negative Gram stain based on moderate evidence acquired through trials (source: Clinical Infectious Diseases, PubMed).

The cerebrospinal fluid analysis model exposed in the Features and General Symptoms of Bacterial Meningitis section that put forth a set of criteria (glucose level below 0.34g/L, cerebrospinal glucose to blood glucose ratio below 0.23, protein level above 2.2g/L or white blood cell count above 2000/mm^3) predictive of bacterial meningitis with 99% certainty, though validated, should not be relied upon to distinguish between bacterial and viral meningitis when it comes to making clinical decisions regarding treatment. There is no agreed-upon test to definitively distinguish between bacterial and viral meningitis in the event of negative cerebrospinal culture and Gram stain results, though a combination of results may be used to weigh the odds of either condition accurately. There are several diagnostic tests used for this purpose (source: Clinical Infectious Diseases, PubMed).

Testing for cerebrospinal **lactate concentration** can help to differentiate between cases of bacterial and aseptic/viral meningitis, as episodes of bacterial meningitis present elevated levels of lactate. In a study of 53 episodes of meningitis, 25 bacterial and 28 viral, the lactate

come into contact with a sample in order to test for the presence of a specific antigen. A positive agglutination test involves the agglutination or clumping together of antigens under the influence of the antibody-coated capsules. Results are normally available within 15 minutes. See fig. 2

57 Negative predictive values: Proportion of patients with negative test results who are correctly diagnosed.

concentration was measured on the initial lumbar puncture before antimicrobial treatment had been issued. The average concentration in episodes of bacterial meningitis was 13.6mmol/L, with a range of 3.5 to 24.5, and the average concentration in episodes of viral meningitis was 2.7mmol/L, with a range of 1.4 to 4.2. Because of the presence of several factors unrelated to bacterial meningitis that have the capacity to elevate lactate concentration, it is not a recommended test to confirm bacterial meningitis (source: Clinical Infectious Diseases, PubMed).

Another diagnostic test that can distinguish bacterial meningitis from viral meningitis is the measurement of **C-reactive protein concentration** in blood serum. In a study published in the Journal of Paediatrics in 1999, the average concentration of C-reactive protein in blood serum was compared between 325 episodes of culture-positive bacterial meningitis and 182 children with proven or presumed viral meningitis. The average concentration in episodes of bacterial meningitis was 115mg/L, and in episodes of viral meningitis, <20mg/L (source: Clinical Infectious Diseases, PubMed).

Cerebrospinal fluid **PCR** can be used to diagnose quickly enteroviral meningitis, the most common form of viral meningitis, and is considerably faster and more sensitive than a viral culture for enteroviruses, which allows for, primarily, shortened hospitalisation for most patients of viral meningitis tested. As well enteroviral meningitis, it can also be used to diagnose herpes simplex, varicella-zoster, arboviruses, Epstein-Barr and mumps viral meningitis (sources: Cecil Medicine, Clinical Infectious Diseases, BMJ, PubMed).

5.1.2 Diagnostic techniques for the detection of viral meningitis

Viral meningitis often goes unreported and is largely benign and free of complications, but, as well as prolonging hospitalisation and prompting the unnecessary use of antibiotic treatment, it can also cause considerable morbidity[58]. The majority of prime viral causes of meningitis are diagnosed primarily through cerebrospinal fluid PCR, but these can also be diagnosed through tests like viral cultures or immunofluorescence[59] (source: BMJ, PubMed).

58 Morbidity: Poor health.

59 Immunofluorescent diagnosis: Any of several methods that use antibodies linked to fluorescent dyes to identify antigens in a sample.

5.2 Treatment of meningitis

5.2.1 Treatment of bacterial meningitis

Given the severity of bacterial meningitis, **empirical treatment** should be administered directly after blood has been drawn and a lumbar puncture has been performed (unless a CT scan was deemed necessary before the lumbar puncture, in which case the empirical treatment should already be underway). If bacterial meningitis is diagnosed and the causative organism is established through a cerebrospinal fluid Gram stain or another diagnostic test, **targeted antimicrobial treatment** can be given based on the pathogen in question. If bacterial meningitis is suspected but the causative organism has not been established, empirical treatment is continued. **Specific antimicrobial treatment** can be given if the causative organism is isolated and in vitro susceptibility testing[60] is performed (sources: Cecil Medicine, Clinical Infectious Diseases, PubMed).

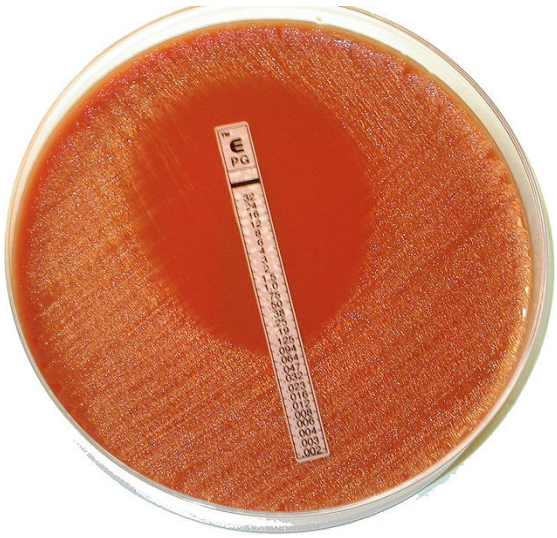

fig. 3 Image depicting a method of susceptibility testing against Neisseria gonorrhoeae: the Epsilometer test strip (or E-test), which contains a gradation of antibiotic, with the strongest at the top and weakest at the bottom. A zone of inhibition wherein the bacteria do not grow starting somewhere on the gradation indicates the smallest concentration required to visibly disturb the colony of bacteria growing on the plate, which correlates with the effectiveness of the antibiotic. Source: Wikimedia Commons.

The recommended empirical treatment is based on age group and other predisposing factors, as well as the initial assumption that antimicrobial resistance is present. In neonates (patients below the age of one month) with suspected bacterial meningitis, the

60 In vitro susceptibility testing: Laboratory trial testing the sensitivity of microorganisms towards potential treatments. See fig. 3 and fig. 4

recommended empirical treatment is ampicillin[61] and cefotaxime[62]/aminoglycoside[63], in all patients between the ages of one month and 50 years the recommended treatment is vancomycin[64] and a third-generation cephalosporin[65], whilst the recommended empirical treatment for patients over the age of 50 is vancomycin and ampicillin as well as a third-generation cephalosporin. Patients who have basilar skull fracture as a predisposing factor are recommended vancomycin and a third-generation cephalosporin, whilst those with penetrating trauma, recent neurosurgery or a cerebrospinal fluid shunt are recommended vancomycin and cefepime[66], ceftazidime[67], or meropenem[68]. All antimicrobial treatment should be administered intravenously. Although targeted antimicrobial treatment is issued for adults based on presumptive identification of the causative bacteria, in infants and children the continuation of empirical treatment is recommended (sources: Cecil Medicine, Clinical Infectious Diseases, PubMed).

If ***S. pneumoniae*** is detected in adults, the recommended antimicrobial treatment is vancomycin (historically, penicillin was the treatment of choice, though the rise of penicillin-resistant pneumococcal strains all around the world prompted its replacement) alongside a third-generation cephalosporin due to the wide distribution of resistant strains. If the penicillin minimum inhibitory concentration[69] (abbreviated MIC) is found to be below 0.1µg/mL, the recommended specific treatment is Penicillin G[70] or vancomycin, if between 0.1 and 1µg/mL, a third-generation cephalosporin, and if above 2µg/mL, or a cefotaxime/ceftriaxone MIC is above 2µg/mL, vancomycin alongside a third-generation

61 Ampicillin: Semisynthetic beta-lactam antibiotic (acts by inhibiting the synthesis of bacterial cell walls) belonging to the penicillin family.
62 Cefotaxime: Member of the third generation of cephalosporins.
63 Aminoglycosides: Group of antibiotics composed of amino sugars united through a glycosidic bond, derived from various species of *Streptomyces* bacteria.
64 Vancomycin: Glycopeptide antibiotic (similarly to beta-lactam antibiotics, acts by inhibiting the synthesis of bacterial cell walls) isolated from cultures of the bacteria *Amycolaptopsis* (formerly *Streptomyces*) *orientalis*.
65 Third-generation cephalosporin: Third group of the broad-spectrum beta-lactam class of antibiotics, cephalosporins, derived historically from the fungi *Cephalosporium acremonium*. Unless indicated, "third-generation cephalosporins" referenced for treatment will be cefotaxime or ceftriaxone, which are considered equivalent antibiotics in terms of efficiency.
66 Cefepime: Fourth-generation cephalosporin.
67 Ceftazidime: Third-generation cephalosporin.
68 Meropenem: Broad-spectrum antibiotic used primarily in the treatment of bacterial meningitis and intra-abdominal infections.
69 Minimum inhibitory concentration: Smallest concentration of antibiotic needed to create a visible inhibition of growth on a culture plate. See fig. 3
70 Penicillin G: Most commonly used penicillin compound, also known as benzylpenicillin.

cephalosporin. The recommended duration of the treatment is between 10 and 14 days (sources: Cecil Medicine, Clinical Infectious Diseases, PubMed).

If ***N. meningitidis*** is detected in adults, the recommended antimicrobial treatment is a third-generation cephalosporin (as with *S. pneumoniae*, *N. meningitidis* strains with penicillin-resistance have been isolated (particularly in Spain) prompting the use of cephalosporins in targeted, non-specialised treatment). If the penicillin MIC is found to be below 0.1µg/mL, the recommended specific treatment is Penicillin G or ampicillin, and if equal to or above 0.1µg/mL, a third-generation cephalosporin. The recommended duration of the treatment is 7 days (sources: Cecil Medicine, Clinical Infectious Diseases, PubMed).

If ***H. influenzae*** is detected in adults, the recommended antimicrobial treatment is a third-generation cephalosporin. If found through susceptibility testing to be beta-lactamase[71] negative, ampicillin is the recommended specific treatment, and if positive, a third-generation cephalosporin. The recommended duration of the treatment is 7 days (source: Clinical Infectious Diseases, PubMed).

If ***S. agalactiae*** is detected in adults, the recommended antimicrobial treatment is ampicillin or penicillin G. There is no susceptibility test required and no recommended specific antimicrobial treatment. The recommended duration of the treatment is between 14 and 21 days. If ***L. monocytogenes*** is detected in adults, the recommended antimicrobial treatment is ampicillin or penicillin G. There is no susceptibility test required and no recommended specific antimicrobial treatment. The recommended duration of the treatment is equal to or above 21 days. If ***E. coli*** or another aerobic gram-negative bacilli is detected in adults, the recommended antimicrobial treatment is a third-generation cephalosporin. There is no susceptibility test required and no recommended specific antimicrobial treatment. The recommended duration of the treatment is 21 days (source: Clinical Infectious Diseases, PubMed).

5.2.2 Treatment of viral meningitis

In some cases of viral meningitis, such as enteroviral, mumps or arbovirus meningitis, there is

71 Beta-lactamase: Bacteria-produced enzyme responsible for resistance to beta-lactam antibiotics like penicillin, though cephalosporins are relatively resistant to it.

no specific antiviral treatment, and only supportive therapy is employed, whereas with others, such as herpes simplex viruses and varicella-zoster virus, there is specific antiviral treatment associated (though not necessarily specific to meningitis), such as aciclovir[72], a standard drug in most first episodes of those viruses (source: BMJ, PubMed).

5.3 Prevention of meningitis

5.3.1 Prevention of bacterial meningitis

Note: all information related to the issuing of vaccinations at certain ages is drawn from the Department of Health of Catalonia and is, therefore, tailored to this region.

Among vaccinations for bacterial meningitis, perhaps none have been as impacting as the vaccination for ***H. influenzae***, which successfully reduced in incidence of *H. influenzae* meningitis by over 94-99% of cases in regions receiving universal HiB vaccination coverage and a decline of cases of community-acquired meningitis (documented in the United States) of 55%. Polyribitol phosphate[73] (abbreviated PRP) vaccinations first became available in 1985, but, due to poor immune responses in infants, these had to be administered to children over the age of 2 years, which did not provide immunization for the most susceptible agegroups. This prompted the development PRP vaccinations conjugated with carrier proteins, which are now administered universally to all children at the ages of 2, 4, 6 and 18 months (sources: Lancet, Sherris Medical Microbiology, DepSalut, Cecile Medicine, PubMed).

There is no vaccine effective against listeriosis (infection by ***L. monocytogenes***), and only methods such as the avoidance of any and all uncooked or unpasteurized foods, unwashed vegetables or prepared foods and any improperly contained perishable foods can be employed in prevention (source: Centers for Disease Control and Prevention).

72 Aciclovir: Antiviral drug used primarily for the treatment of infections stemming from the most well known species in the herpesvirus family.

73 Polyribitol phosphate: Capsule of the type b strain of *H. influenzae*, which is composed of a polymer of ribose, ribitol and phosphate.

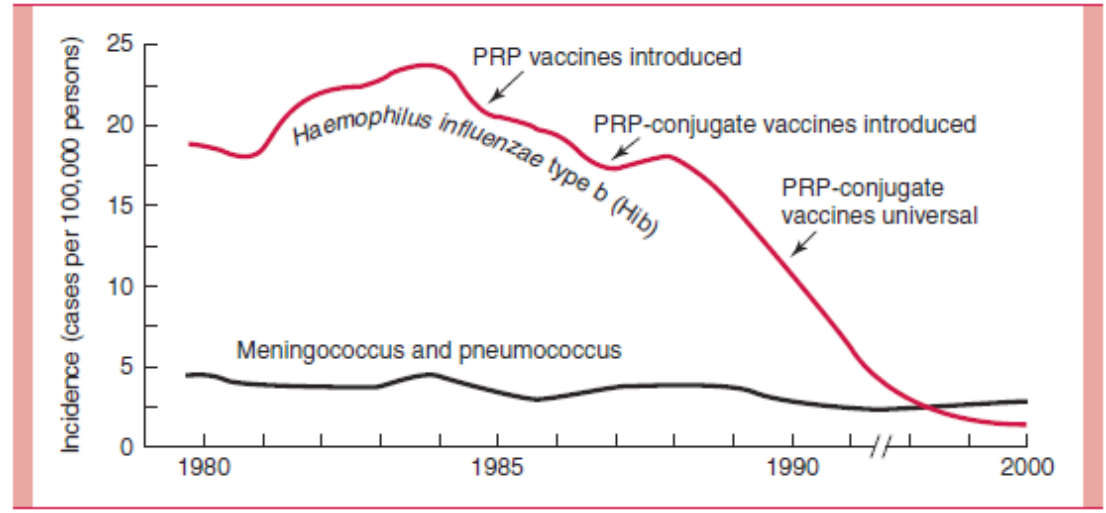

fig 5. Image depicting the decline in cases of H. influenzae type b meningitis in the United States in relation to the introduction of new vaccines. Source: Sherris Medical Microbiology.

The preventative methods that can be used against infection through ***E. coli*** are similar to those employed to stave off listeriosis: eating only cooked foods and peeled fruits and drinking only hot or carbonated beverages, avoiding raw vegetables and uncertain water especially when travelling in developing countries. There are no widespread approved vaccines for *E. coli*, but there are some in development against particular strands of the bacteria (sources: Sherris Medical Microbiology, Centers for Disease Control and Prevention).

Various approved vaccinations are available against different serotypes of ***N. meningitidis***: two multivalent[74] vaccines, one against serotypes A, C, Y and W-135 and the other against A and C, are available for selective vaccination for individuals travelling to regions where epidemics of these serotypes are common, and one monovalent vaccine against meningococcus C, which is issued to children at the ages of 2, 6 and 15 months. The development of a vaccination immunising against meningococcus serotype B is underway and being used in infant clinical trials. As mentioned in the prime causes of meningitis section of the project, meningococcus is spread through respiratory droplets, but can be, depending on the strain and susceptibility, contagious, in which case, preventative methods would include the screening for meningococcal infections in all individuals who have come into contact with a patient with confirmed meningococcal meningitis (sources: Sherris Medical Microbiology, DepSalut, BMJ).

74 Valency: When pertaining to vaccinations, valency indicates the number of strains or serotypes of a disease a vaccine protects against.

Biology

There are two primary anti-**pneumococcal** approved vaccinations available, a 7-valent vaccine of serotypes 4, 6B, 9V, 14, 18C, 19F and 23F and a 23-valent vaccine of serotypes 1-5, 6B, 7F, 8, 9N, 9V, 10A, 11A, 12F, 14, 15B, 17F, 18C, 19A, 19F, 20, 22F, 23F and 33F. The former vaccination is being considered for use in all children under the age of 5, and both are used selectively in risk-groups, such as those individuals of advanced age, or with conditions such as hemoglobinopathies, asplenia[75], immunodeficiencies, renal insufficiency, chronic respiratory diseases, Down-syndrome, malignant neoplasties (source: Sherris Medical Microbiology, DepSalut).

Although there is no vaccine effective against **GBS** in circulation, and its mechanism for transmission between non-pregnant adults and older children is unknown, pregnant women can be screened for the bacteria through a vaginal and rectal culture between the 35th and 37th weeks of pregnancy. If the culture is tested positive for GBS, antibiotics can be administered intravenously during labour to effectively prevent transmission of the bacteria to the newborn (source: Centers for Disease Control and Prevention).

5.3.2 Prevention of viral meningitis

There are no vaccines for the causative agents of **enteroviral** meningitis (non-polio enteroviruses) or effective targeted antiviral treatments available, limiting all care to supportive treatment, and the only real method of prevention, due to the majority of patients who experience enteroviral infections asymptomatically and its transmission through fecal matter, is general hygiene (sources: Centers for Disease Control and Prevention, Sherris Medical Microbiology).

Highly effective **mumps virus** vaccines have been available since 1967, which has rendered the infection relatively rare. The vaccine for mumps is commonly combined with the vaccine for measles and rubella (the resulting vaccine named MMR) and administered at 12 months and again at 4 years of age, providing potentially lifelong immunity (sources: Sherris Medical Microbiology, DepSalut).

75 Asplenia: Lack of normal spleen function.

Biology

There are no vaccines for **herpes simplex** or **Epstein-Barr** viruses, though there is an approved long-term immunity vaccination for **varicella-zoster** virus (first synthesized in 1974), which is administered at 12 years of age to all children who have not experienced the disease. The vaccine can also prevent the disease within 3 days after exposure to VZV, which is extremely contagious. Methods of prevention against herpes simplex viruses are limited to avoiding contact with individuals with lesions, though the virus may be carried asymptomatically. As for EBV, vaccinations are under exploration for use in endemic areas and for certain manifestations of the virus. Treatment for it is largely supportive, as targeted antiviral drugs (acyclovir) have not been shown to impact the clinical course of the disease (sources: Sherris Medical Microbiology, Centers for Disease Control and Prevention, DepSalut).

There is no reportedly effective specific treatment available for **arboviruses**, other than supportive treatment. The only arbovirus vaccine available for general human use is the yellow fever virus (a flavivirus) vaccine, which is distributed to rural populations in endemic areas and international travellers. Vaccinations are available for horses against infection from eastern and western equine encephalitis viruses. Prevention of infection is limited to the control of affected arthropod vector, sometimes through the elimination of breeding sites (source: Sherris Medical Microbiology).

The usual treatment for cryptococcal disease (originated by ***C. neoformans***) is amphotericin B[76] or fluconazole[77], and although approximately three quarters of patients with cryptococcal meningitis respond to the treatment, a significant quantity relapse when the antifungal treatment is stopped. Chronic cryptococcosis requires repeated courses of such treatment. As with cryptococcal disease, candidiasis (originated by ***C. albicans***) usually responds to amphotericin B and other antifungals (source: Sherris Medical Microbiology).

76 Amphotericin B: Intravenous antifungal drug employed in cases of systemic fungal infections.
77 Fluconazole: Antifungal drug employed in cases of superficial or systemic fungal infections.

6. Epidemiology of bacterial meningitis

An evaluation of the epidemiology of bacterial meningitis in Catalonia through data catalogued by the Microbiological Notification System of Catalonia (*Sistema de Notificació Microbiològica de Catalunya*) or SNMC from the year 1994 to 2009, as well as data from the Vigilance and Response to Public Health Emergencies General Subdirective (*Subdirecció General de Vigilància i Resposta a Emergències de Salut Pública*) from the year 2000 to 2009, all published in the Epidemiological Bulletin of Catalunya (*Butlletí Epidemiològic de Catalunya*). In four of the sixteen years of examined data (2002, 2003, 2008 and 2009), age group classification of observed cases of bacterial meningitis was absent, as were some bacteria and serological classifications covered in this project, which prompted their exclusion from several data ranges.

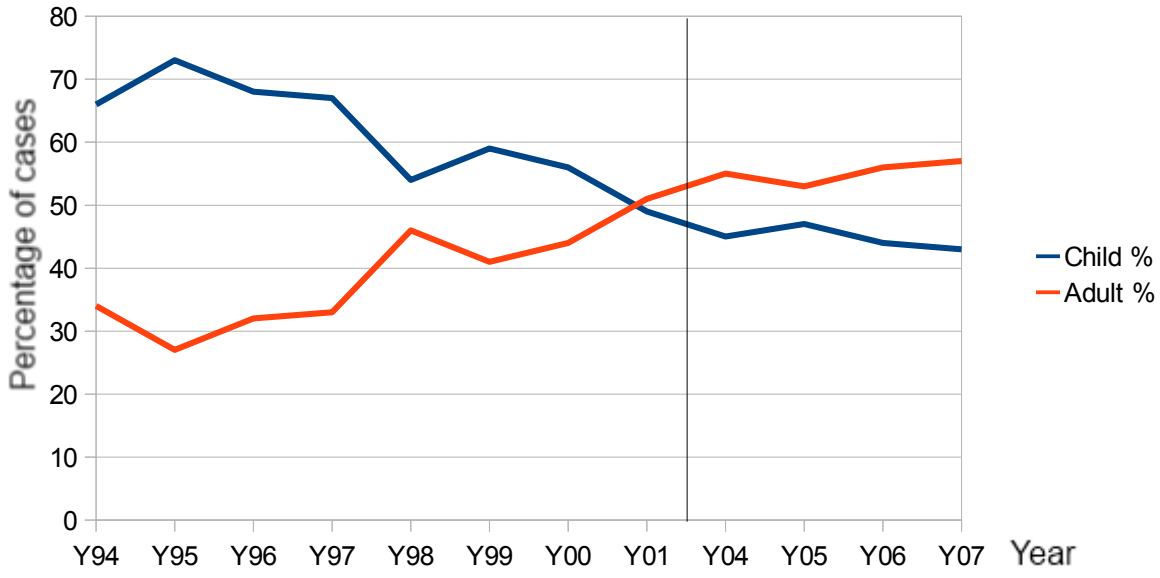

fig. 1 Graph detailing the proportions of bacterial meningitis in Catalonia in adults and children by year. *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

Though in the first seven documented years the recorded cases of childhood bacterial meningitis (defined here as bacterial meningitis afflicting patients equal to or below the age of 19) outnumber those of adults, the proportion of adult bacterial meningitis increases to

overtake that of child bacterial meningitis.

Together, meningococcus and pneumococcus have accounted for between 72 and 94% of cases of bacterial meningitis. In order to distinguish between a rise of overall cases of pneumococcal meningitis, a decrease in overall cases of meningococcal meningitis, or both, the cases drawn by the Microbiological Notification System of Catalonia can be consulted. As time progresses, more laboratories are added to the pool of the Microbiological Notification System of Catalonia, starting with 18 in 1994 and building up to 40 in 2009. However, taking this into account, the comparison of a range of 88-147 cases of meningococcal meningitis per year between 1994 and 2000 with a laboratory range of 18-31 versus a range of 40-91 cases per year between 2001 and 2009 with a laboratory range of 33-40 indicates an overall decrease in cases of meningococcal meningitis.

This is corroborated by the overall decrease examined in cases of invasive meningococcal disease recorded between 2000 and 2009, which is an obligatory declaration disease, and is, therefore, not biased in terms of laboratory count. As can be observed in figure 3, the graph of total meningococcal disease cases per year closely resembles the graph of meningococcal

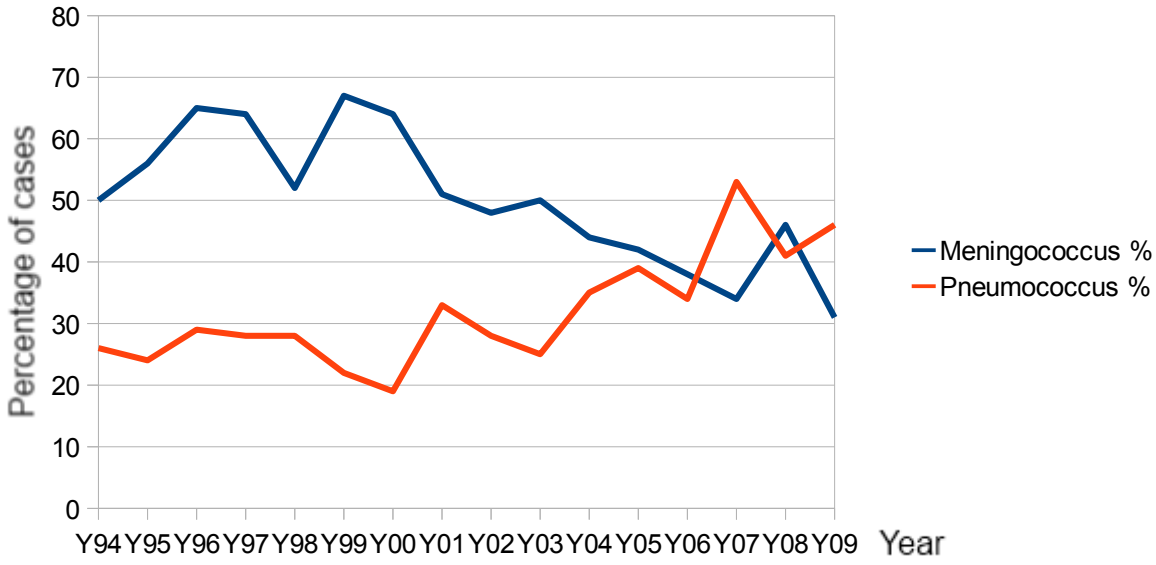

fig. 2 Graph detailing the percentage of cases of bacterial meningitis in Catalonia caused by meningococcus and pneumococcus by year. Years 2002 and 2003 have more approximated values. Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.

meningitis based on the Microbiological Notification System of Catalonia, and is based on data collected through a completely different system, the obligatory disease declaration system.

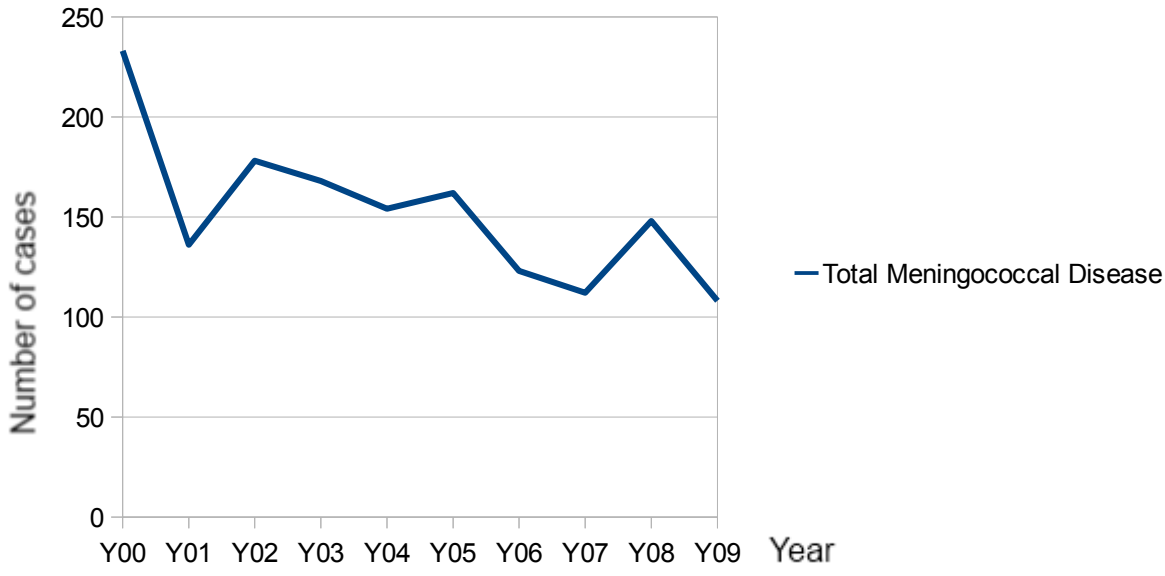

fig. 3 Graph representing overall cases of meningococcal disease in Catalonia by year, showing the gradual decline of infection caused by N. meningitidis. *Source: Own work, based on data collected by the Subdirecció General de Vigilància i Resposta a Emergències de Salut Pública and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

6.1 *Streptococcus pneumoniae*

S. pneumoniae is, following the most recent publication regarding microbiological organism distributions, the most common cause of bacterial meningitis in Catalonia. The incidence of pneumococcal meningitis is most prominent in the over-59 age group, and there has only been one year in which more cases of pneumococcal meningitis have been attributed to another age group (in 2005, to the 40-49 age group). Neonatal meningitis is rarely attributed to pneumococcus, but excepting that, *S. pneumoniae* can be suspected as the causative organism of bacterial meningitis in all other age groups, with the over-30 age group being the most common.

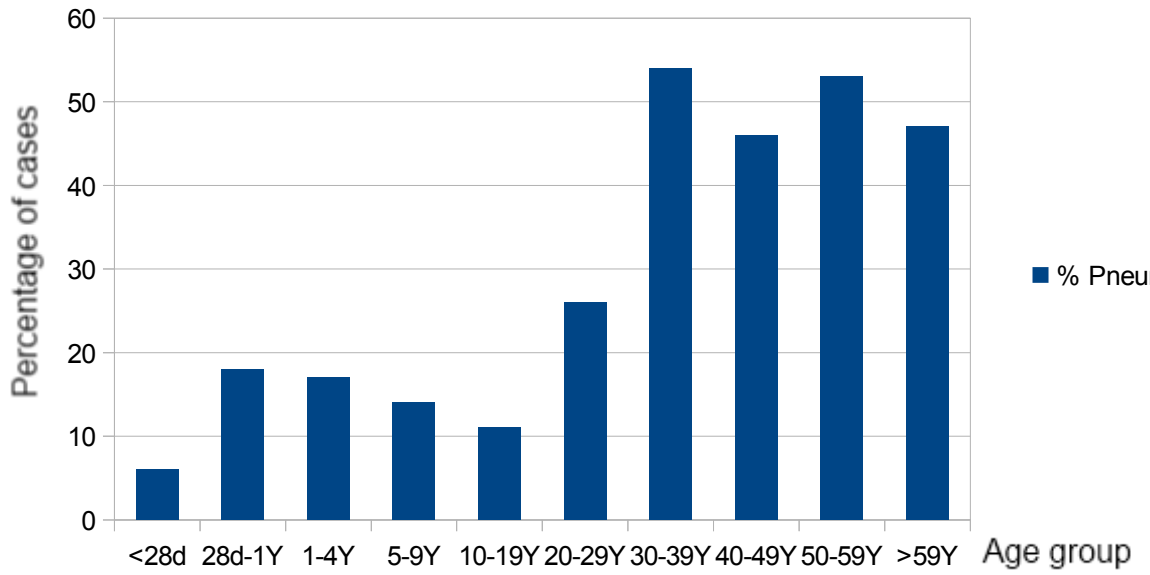

Fig. 4 Graph depicting overall occurrence of pneumococcal meningitis in Catalonia by age group as a causative agent from 1994 to 2008 (excluding 2002-03 and 2008-09). *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

Though the over-59 age group is the focus of *S. pneumoniae* by far, pneumococcus has a similar occurrence in the 30 to 59 age groups, indicating a heightened number of cases of bacterial meningitis caused by other organisms in the over-59 age group competing with pneumococcus, but not in the 30-59 age group.

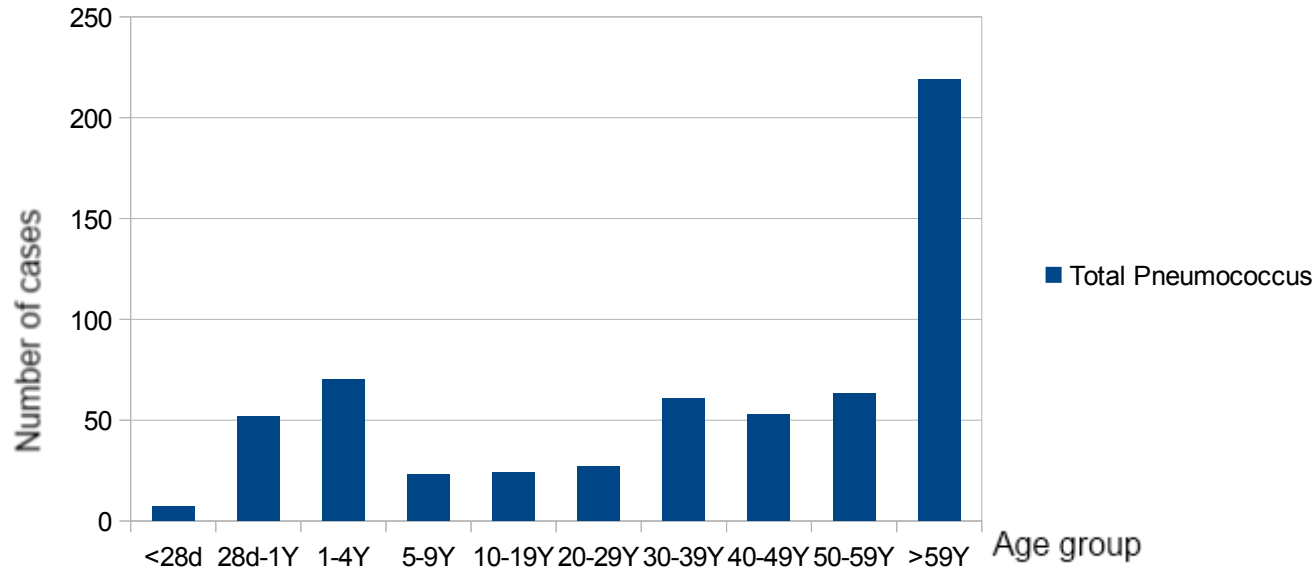

fig. 5 Graph depicting overall cases of pneumococcal meningitis in Catalonia by age group, highlighting the focus of most cases in the over-59 age group. *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

6.2 *Neisseria meningitidis*

N. meningitidis has been, in all but two years (2007 and 2009) of the examined timeframe, the most prominent causative agent of bacterial meningitis. Unlike S. pneumoniae, the majority of cases of meningococcal meningitis are found in the 28 days to 19 years age groups, which have accounted for the majority of all cases of bacterial meningitis in those age groups in all documented years.

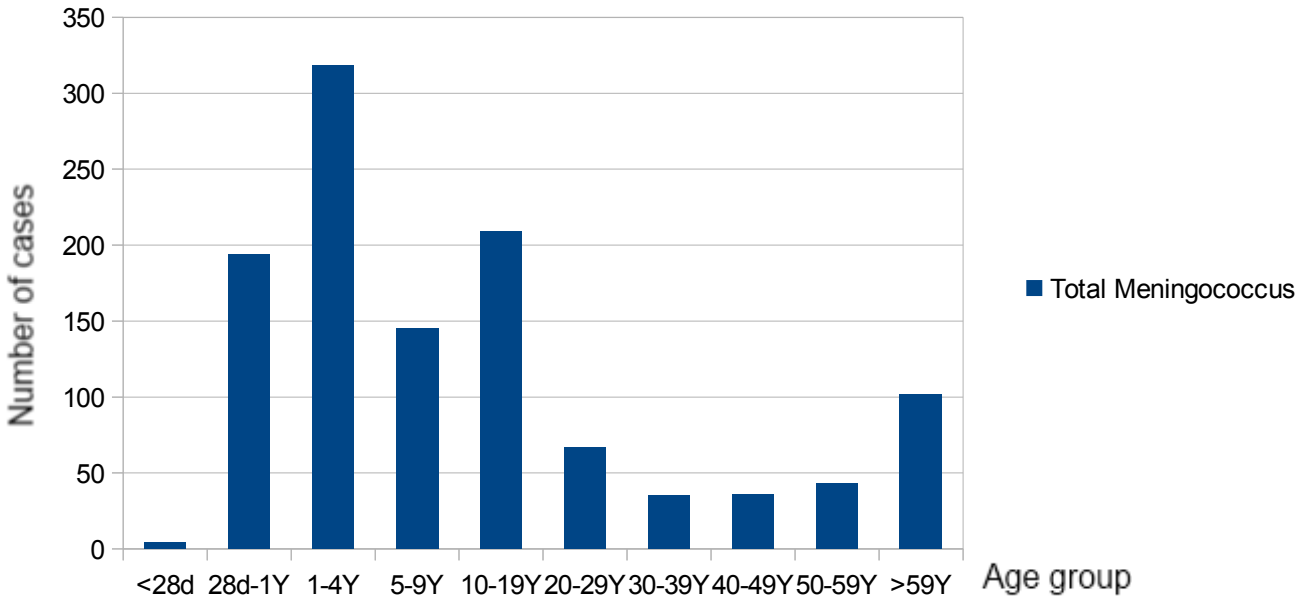

fig. 6 Graph depicting overall cases of meningococcal meningitis in Catalonia by age group, depicting the focus of most cases in post-neonatal child age groups from 1994 to 2008 (excluding 2002-03 and 2008-09). Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.

As with S. pneumoniae, neonatal meningitis is rarely attributed to N. meningitidis, and meningococcus can be suspected as the causative organism in all other age groups. Of the examined serotypes of meningococcus, serotype B accounts for the majority of all cases of meningococcal meningitis in all years, followed by serotype C, both experiencing a decline in occurrence because of the overall decline of cases of meningococcal meningitis.

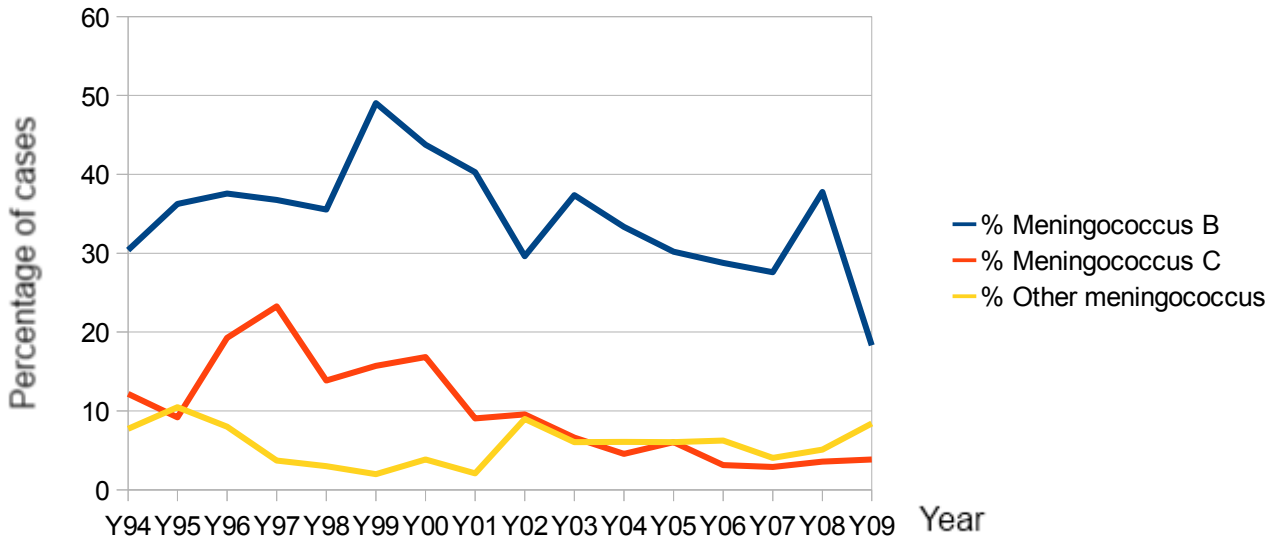

fig. 7 Graph detailing the percentage of cases of bacterial meningitis in Catalonia caused by different serological groups of meningitis by year. *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

Even though the great majority of cases of meningococcal meningitis are spread across the 1 month to 19 years age groups, it also presents as the highest cause of some adult age groups, such as 20-29 and 40-49. This is due to its overall occurrence rate as a causative organism throughout the examined timeframe.

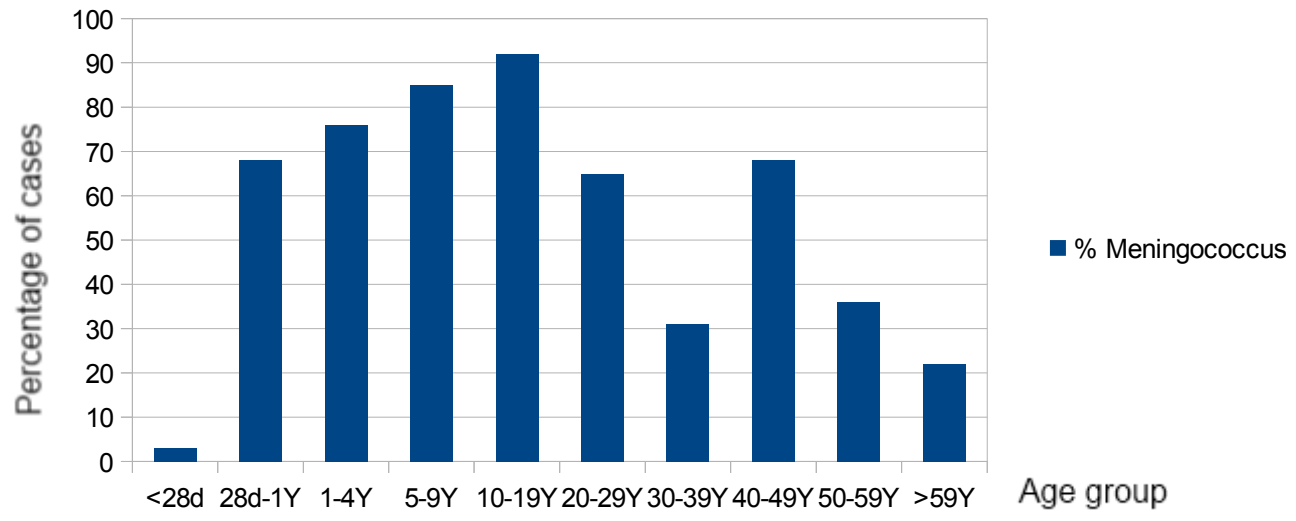

fig. 8 Graph depicting overall occurrence of meningococcal meningitis in Catalonia by age group as a causative agent from 1994 to 2008 (excluding 2002-03 and 2008-09). *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

6.3 *Streptococcus agalactiae*

S. agalactiae has, throughout the examined timeframe, been the most prominent cause of meningitis in neonates (defined as children below the age of 28 days), with the exception of two years (2004 and 2007). Cases of neonatal meningitis have always accounted for the greater majority of episodes of meningitis induced through GBS, with the exception of one year which presented only one case of meningitis caused by GBS.

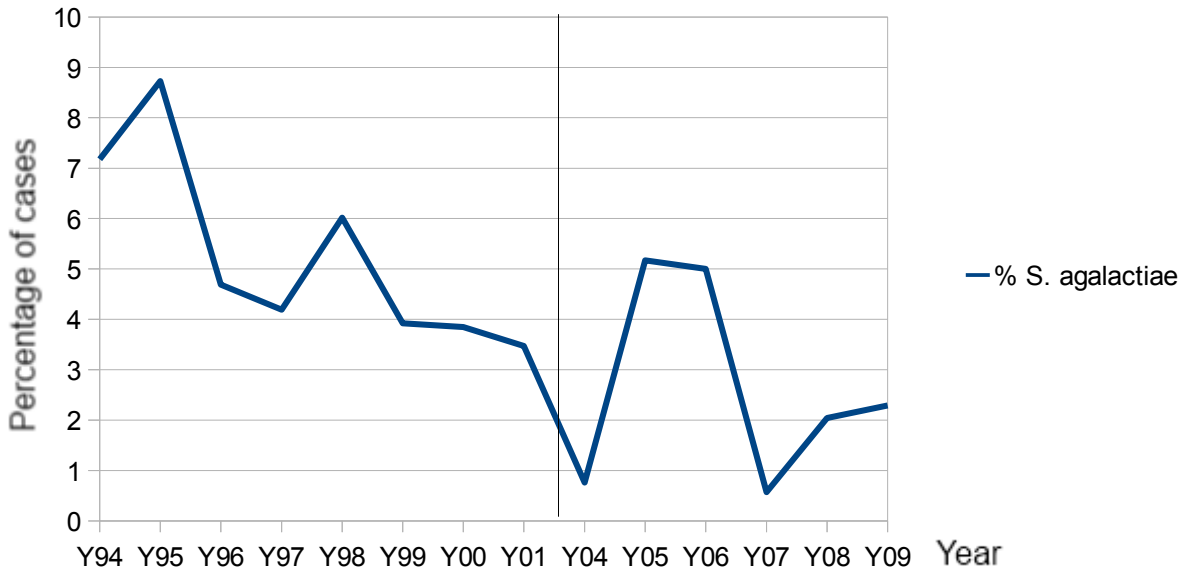

fig. 9 Graph depicting the occurrence of S. agalactiae among cases of bacterial meningitis in Catalonia and showing its gradual decline as a causative agent. *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

S. agalactiae is responsible for 70% of all cases of neonatal meningitis in the examined timeframe (all sixteen years excepting the four in which age group classification was absent), and its decline correlates with the decline of the occurrence of neonatal meningitis shown below.

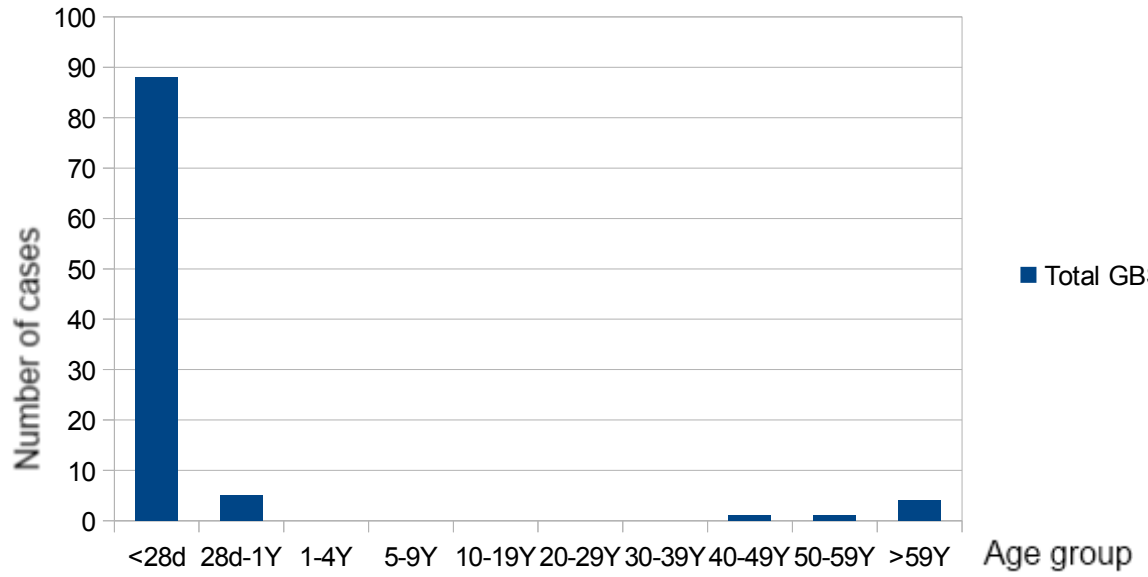

fig. 10 Graph depicting overall cases of meningitis caused by S. agalactiae in Catalonia by age group, highlighting the focus of nearly all cases in the neonatal age group from 1994 to 2008 (excluding 2002-03 and 2008-09). *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

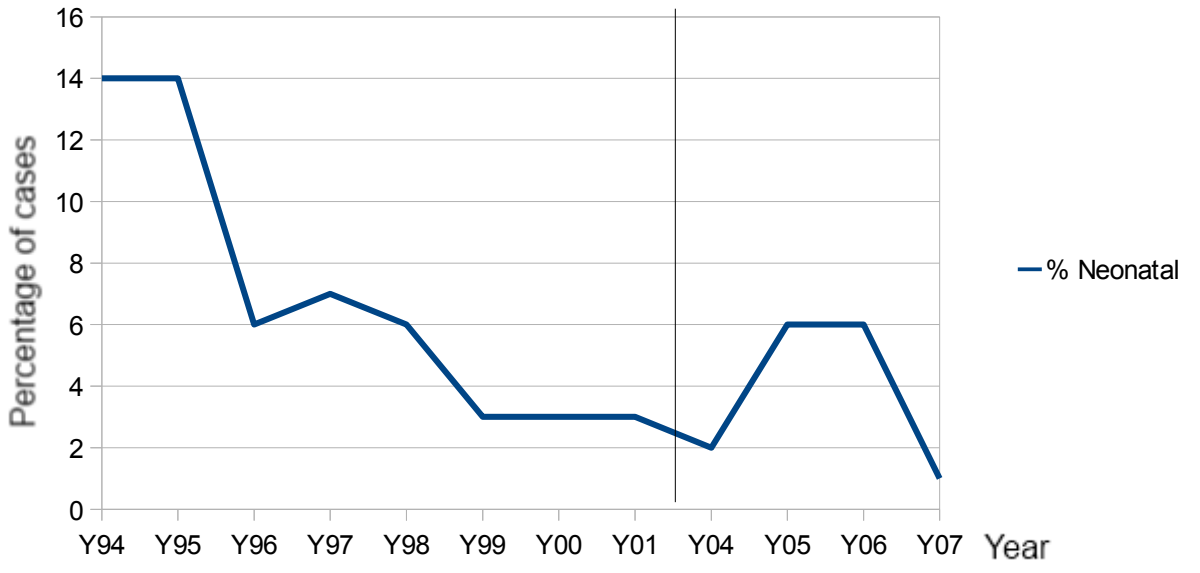

fig. 11 Graph representing the overall decline of the occurrence of neonatal bacterial meningitis in Catalonia. *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

6.4 *Listeria monocytogenes*

L. monocytogenes is found primarily in patients over the age of 59, where most if not all of its isolations have been centred in with the exception of two years, as well as in neonates and in the 30 to 59 age groups, though to a lesser extent. There can be rare occurrences in which the bacterium is isolated in patients between the ages of 28 days and 30 years.

fig. 12 Graph depicting the occurrence of L. monocytogenes among cases of bacterial meningitis in Catalonia, showing its gradual ascent as a causative agent. Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.

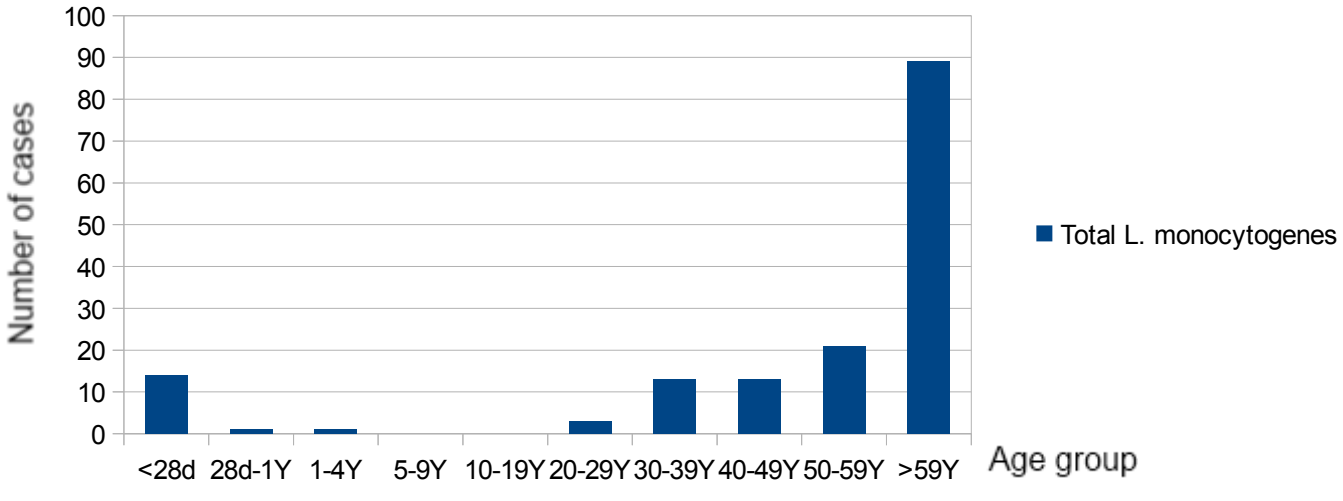

fig. 13 Graph depicting overall cases of listeria meningitis in Catalonia by age group, highlighting the focus of the majority of all cases in the over-59 age group from 1994 to 2008 (excluding 2002-03 and 2008-09). Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.

6.5 *Haemophilus influenzae*

The rate of recorded cases of ***H. influenzae*** meningitis experiences a decline, even with the increased number of laboratories participating in the SNMC program. In the first few examined years, cases of *H. influenzae* serotype B meningitis cluster in the 28 days to 4 years age groups, but as the case numbers drop through the years, episodes are more frequent in adults.

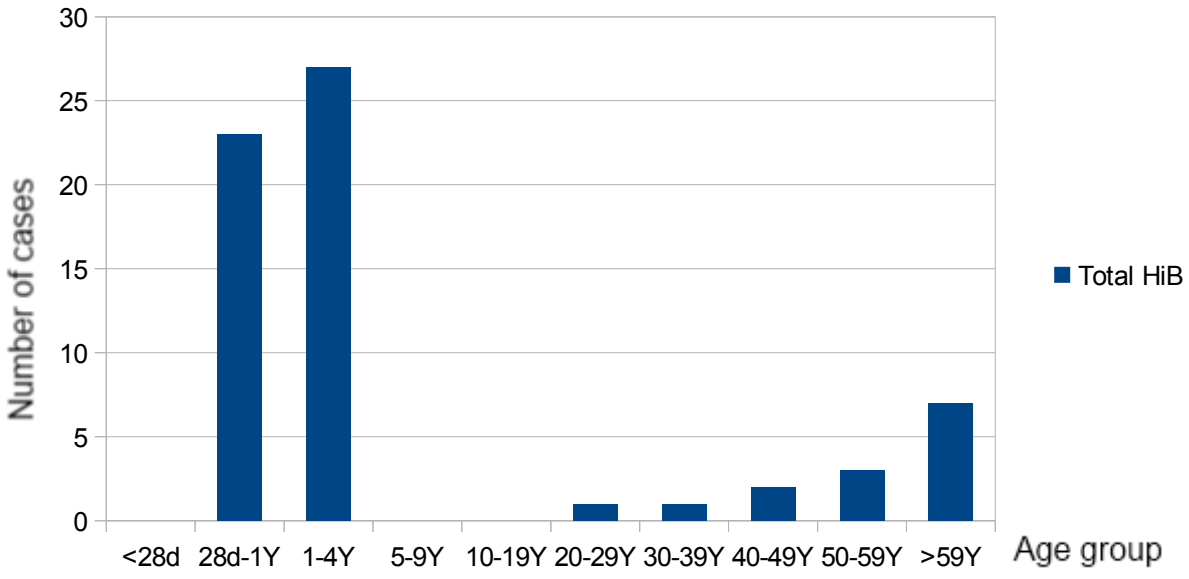

fig. 14 Graph depicting overall cases of meningitis caused by H. influenzae serotype B in Catalonia by age group, depicting the focus of the great majority of cases in post-neonatal child age groups from 1994 to 2008 (excluding 2002-03 and 2008-09). *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

The above graph indicates the infant and child age group focuses of H. influenzae serotype B, which is the most prominent serotype encountered in regions without vaccination coverage of it.

6.6 International research: *H. influenzae* vaccination

The lack of regional *H. influenzae* serotype B immunisation is an important etiological factor, as shown in the partial etiology[78] below. The partial etiology was assembled between 1996

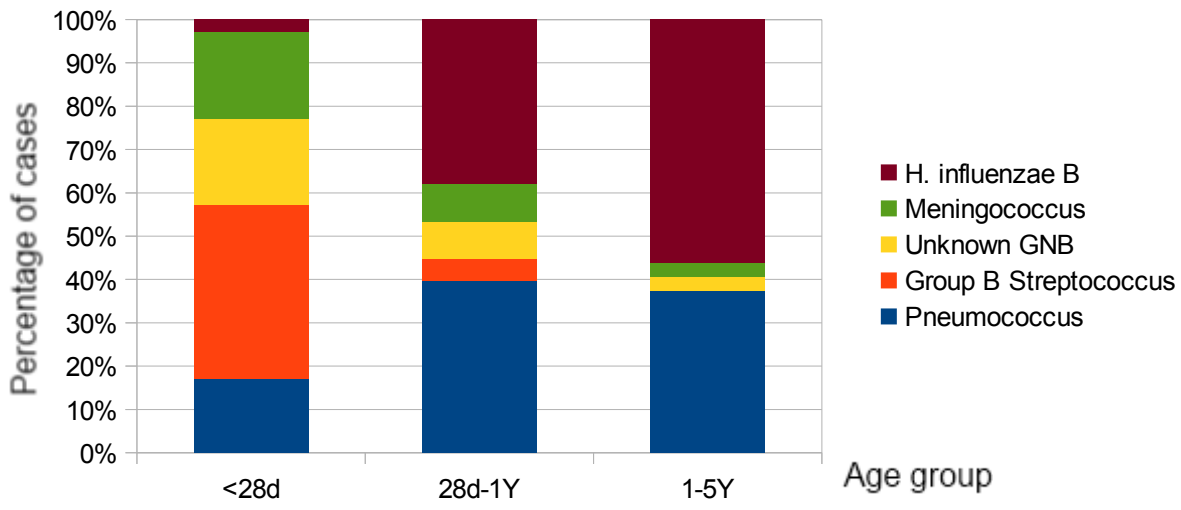

fig. 15 Image depicting the partial etiology of bacterial meningitis in child age groups established through a 1996-1997 study in Malawi, a region with no conjugate HiB vaccine at the time. Source: Adapted from a 1997 study published in the Tropical Medicine and International Health journal.

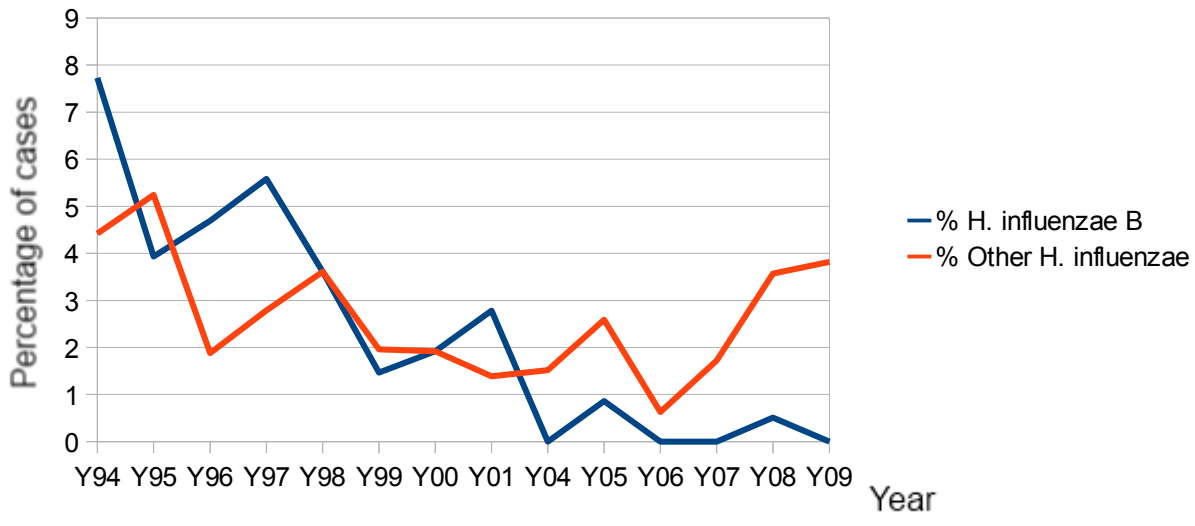

fig. 16 Graph depicting the occurrence of H. influenzae among cases of bacterial meningitis in Catalonia and showing the gradual decline of H. influenzae serotype B. Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.

78 Etiology: Distribution of causes of a disease. In this case, the distribution of causative organisms of recorded cases of bacterial meningitis.

and 1997 in Malawi, approximately five years before the 2002 distribution of a conjugate vaccine immunising against HiB, and represents the prominence of HiB as a causative agent in child age groups.

As shown above, *H. influenzae* B meningitis represents a considerable percentage of all cases of child meningitis in regions without vaccination coverage, in age groups corresponding to the focuses shown in figure 14. Shown below is the collapse in occurrence of H. influenzae serotype B after the introduction of conjugate HiB vaccines in the 1990s.

6.7 International research: epidemiological comparisons

A study done in the United States in 1995 and published in the New England Journal of Medicine in 1997 establishes an etiology for bacterial meningitis based on a surveillance in twenty-two counties in four states (Georgia, Tennessee, Maryland and California) for *N. meningitidis*, *H. influenzae*, GBS, *L. monocytogenes* and *S. pneumoniae* in 1995, which identified 248 patients. The areas in which surveillance was performed cover east coast, west coast and inland counties, so that recorded cases effectively established a general epidemiology for the entire United States.

Approximately 25% (62) of cases of bacterial meningitis were caused by *N. meningitidis*, 47% (117) by *S. pneumoniae*, 12% (31) by *S. agalactiae*, 8% (20) by *L. monocytogenes* and 7% (18) by *H. influenzae*. The age group distribution of these agents varied similarly to those isolated in 1995 by the Microbiological Notification System of Catalonia, though presented some differences, most notably a higher occurrence of pneumococcus and a lower occurrence of meningococcus. In the neonatal period, the most prominent causative agent was GBS, as with the data found in Catalonia, though in infants between the ages of 1 and 12 months, *S. pneumoniae* was encountered more frequently than *N. meningitidis* accounting for 45 and 31% of cases in that age group, respectively.

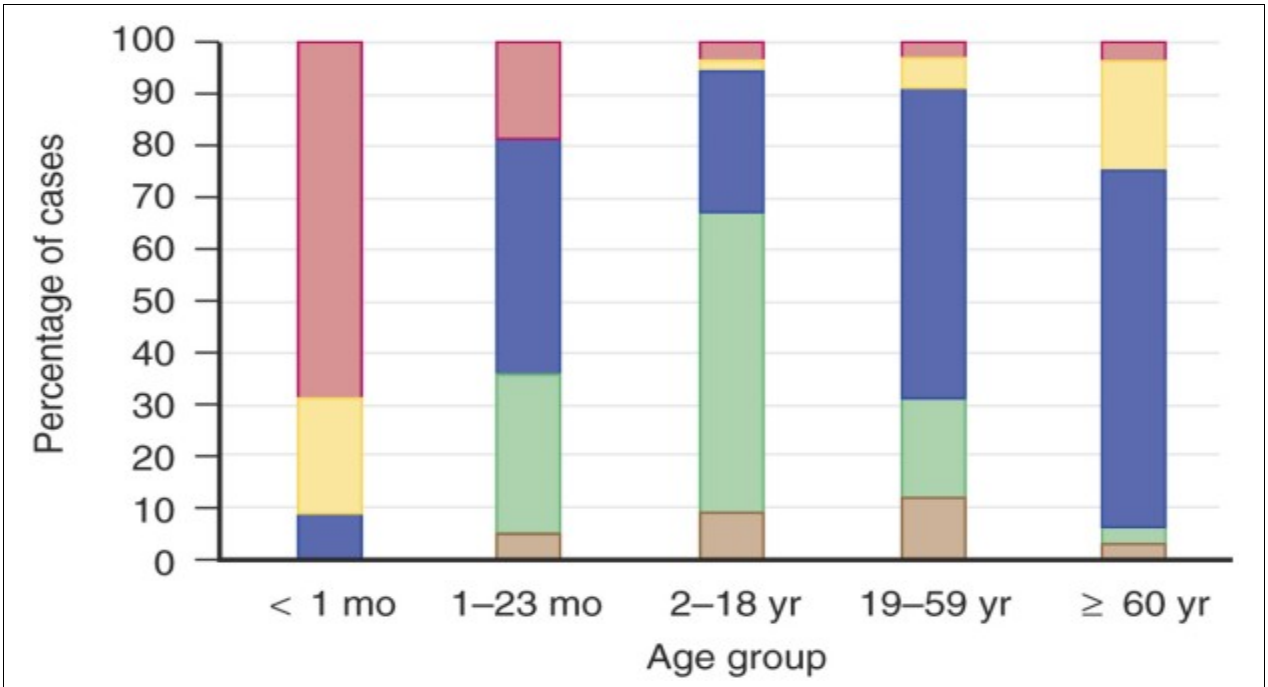

fig. 17 Scanned image depicting the etiology of bacterial meningitis in the United States in 1995. Red = GBS; yellow = L. monocytogenes; blue = S. pneumoniae; green = N. meningitidis; brown = H. influenzae. *Source: New England Journal of Medicine, Cecil Medicine.*

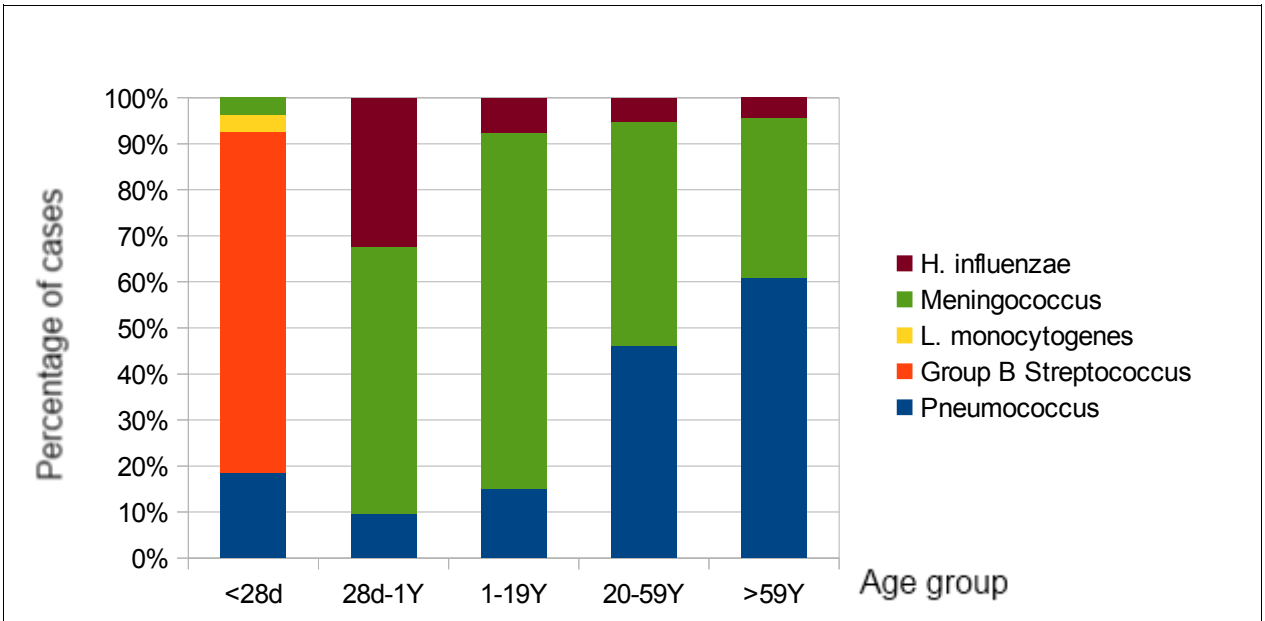

fig. 18 Image depicting the etiology of bacterial meningitis in Catalonia in 1995. *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

Comparing the 1995 United States etiological graph with the 1995 Catalonia one reveals several differences: Catalonia presented a much higher occurrence of meningococcal meningitis across all age groups, a lower occurrence of pneumococcal meningitis across the majority of all age groups, a lower occurrence of *L. monocytogenes* meningitis and a higher occurrence of *H. influenzae* meningitis in children, where the United States graph presented a higher occurrence of *H. influenzae* meningitis in adults.

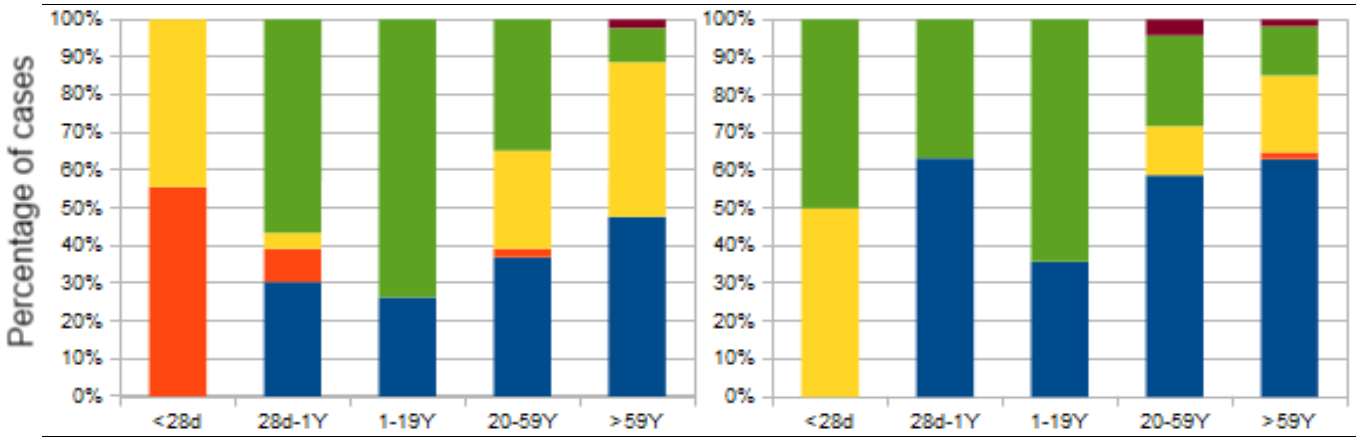

fig. 19 Image depicting the etiology of bacterial meningitis in Catalonia in 2006 (left) and 2007 (right). Orange = GBS; yellow = L. monocytogenes; blue = S. pneumoniae; green = N. meningitidis; brown = H. influenzae. *Source: Own work, based on data collected by the Microbiological Notification System of Catalonia and released in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.*

Subsequently comparing the 1995 Catalonia etiological graph with the 2006 and 2007 ones indicates the shift in causative organism distribution that's since occurred: the occurrence of meningococcal meningitis has decreased, the occurrence of pneumococcal meningitis has increased to fill the gap left by meningococcal meningitis, there is a far higher occurrence of meningitis caused by *L. monocytogenes* and *H. influenzae* meningitis presents a much lower occurrence, now in adults.

The modern distribution of neonatal meningitis in Catalonia can be seen to vary excessively between the 2006 and 2007 graphs because of a low number of recorded cases of it, in comparison to those recorded in 1995 (9 and 2, for 2006 and 2007, versus 30, for 1995).

Biology

The partial etiologies below serve to compare the adult etiology of bacterial meningitis in Catalonia from 1999 to 2001 of the the adult etiology of bacterial meningitis yielded from the 1998-2002 (started in late 1998 and finished in early 2002) study in the Netherlands.

In order to compare the etiological breakdown of the 1998-2002 study conducted in the Netherlands and published in the New England Journal of Medicine, the occurrence of the top five causative agents established over the 4-year study were placed in one column and compared with the occurrence of the same five causative agents established in three separate years of the same time period (1999-2001). The average occurrence of the five causative organisms was then calculated and displayed beside the Netherlands study column to facilitate an epidemiological comparison of the two geographic locations during the same time period.

The overall causative organism breakdowns are compared in the table below.

% Causative Agent	1999 Cat.	2000 Cat.	2001 Cat.	Average Cat.	1998-02 Netherlands
N. meningitidis	52	43	45	47	39
S. pneumoniae	38	31	35	35	53
L. monocytogenes	7	18	15	13	5
H. influenzae	3	7	4	5	2
S. agalactiae	0	1	1	1	1

fig. 20 Table depicting a partial etiology of bacterial meningitis in Catalonia in adult age groups in 1999, 2000 and 2001, as well as an average of the three years, to compare directly with the percentage yielded by the Netherlands study. Note: percentages may not total 100 due to rounding. Source: Own work, based on data collected by the Netherlands Reference Laboratory for Bacterial Meningitis and compiled in the New England Journal of Medicine as well as data collected by the Microbiological Notification System of Catalonia and published in the epidemiological bulletin Butlletí Epidemiològic de Catalunya.

The two most important differences are those observed in the comparison of the 1995 United States and Catalonia etiologies, the higher occurrence of meningococcus and lower occurrence of pneumococcus in Catalonia, though to a smaller extent.

7. Conclusions

During the course of this project I have come to many conclusions surrounding the subject of meningitis, each pertaining to an aspect of the disease I was completely unaware of at the start of this research project.

- The type of meningitis that causes a state of alarm is bacterial meningitis, due to its elevated mortality rate (approximately one fifth of all patients who contract bacterial meningitis die, even with antibiotic treatment supplied) and the likelihood of it causing permanent disability (another fifth of all patients suffer a disability ranging from mild to severely crippling), though viral meningitis is far more commonly encountered.
- Even though viral meningitis can cause short-term disability, almost all cases of it progress with no complications whatsoever and a full recovery. However, complications of viral meningitis can cause permanent disabilities and can even become life-threatening under extreme circumstances.
- Fungal meningitis can prove to be more serious than bacterial meningitis, commonly causing chronic meningitis in those who contract it (and, therefore, increased risk of disability or death), though is far rarer, with some level of immunodeficiency being present in most patients of the disease.
- In comparison with the other types of meningitis discussed, non-infectious meningitis boasts of little research, and its mechanisms are relatively unknown.

- Diagnosing either bacterial or viral meningitis cannot be done based on clinical symptoms alone, as both diseases present the same core symptoms (fever, headache, neck stiffness, change in mental status), both present similar ranges of more uncommon symptoms, and the risk of misdiagnosing bacterial meningitis is too high.

    - Classical symptom-based diagnostic methods, such as identifying Kernig's sign or Brudzinski's sign, have been shown to be unreliable.

- Because of the danger posed by bacterial meningitis, not even the fastest laboratory tests (such as a Gram stain, which can take less than fifteen minutes) can be conducted before beginning some type of antibiotic therapy.
    - It is often possible to distinguish with some confidence between a viral infection of the meninges and a bacterial one based on the colouration of the cerebrospinal fluid drawn from the lumbar puncture, which can allow for empirical antibiotics to be postponed in favour of a diagnostic technique for confirming viral meningitis.
    - Broad-spectrum, empirical antibiotics will have to be administered often due to the causative organism being unknown before laboratory testing can be performed.
- The resistance of certain causative agents of bacterial meningitis towards some broad-spectrum antibiotics makes antibiotic susceptibility testing a necessity, as well as the combination of several broad-spectrum antibiotics for use in empirical antimicrobial therapy.

- The epidemiology of bacterial meningitis has shifted during the last 16 years from one in which the child proportion of cases of bacterial meningitis outnumbered that of the adult one to one in which the adult proportion outnumbers that of the child one.
- This epidemiological shift has been brought about by a gradual decline in cases caused by pathogens with main focuses in child age groups and an increase in cases caused by pathogens with main focuses in adult age groups.
- The most important bacterial pathogens whose alterations in occurrence produced this epidemiological shift were meningococcus and pneumococcus, together

accounting for approximately three quarters or more of cases of bacterial meningitis in all examined years.

- Of the meningococcal serotypes, serotype B accounts for the majority of cases in the developed world. Whilst vaccinations are available for all other serotypes and the development of a vaccination is underway, there is no systematic immunisation program established against meningococcus serotype B.
- A decline in the overall number of cases of meningococcal meningitis is strongly suggested by the data analysed in this section, and the breakdown of occurrence by serotype indicates that the occurrence of all serotypes is decreasing, including that of meningococcus serotype B.
  - The most probable explanation for this decrease, regardless of the lack of vaccinations for serogroup B, lies with the immunisation against other serogroups, or a potential mutation in the organism itself. Source: Dr. Delia Garcia Pares, Head of Infectious Diseases, Hospital Josep Trueta.

- The occurrence of neonatal meningitis has gradually decreased over the last 16 years, correlating with the gradual decrease in occurrence of *S. agalactiae*.
  - This effect can be attributed to the increased usage of intravenous prophylaxis during the labour of women tested positive for GBS (or those displaying signs of infection at the time of labour) during birth. Source: Dr. Delia Garcia Pares, Head of Infectious Diseases, Hospital Josep Trueta.
- The occurrence of meningitis caused by *L. monocytogenes* has gradually increased over the last 16 years.
  - This observed effect can be caused by an increased sensitivity towards recognising cases of *L. monocytogenes* (through the implementation of new and revised diagnostic protocols not present in the past). Source: Dr. Delia Garcia Pares, Head of Infectious Diseases, Hospital Josep Trueta.
- Catalonia has presented a relatively high occurrence of meningococcal meningitis and *L. monocytogenes* meningitis and a relatively low occurrence of pneumococcal meningitis in the years its etiology was compared with the etiologies of other

developed regions (the United States and the Netherlands).

  - This effect is explained by geographical differences, which cause the occurrence of certain types of bacterial meningitis to vary from country to country. Source: Dr. Delia Garcia Pares, Head of Infectious Diseases, Hospital Josep Trueta.

- The HiB conjugate vaccine (administered to infants at 2, 4, 6 and 18 months of age) is very effective in reducing the incidence of HiB meningitis, whereas the standardised pneumococcal vaccine has yet to create a noticeable effect on the present etiology of bacterial meningitis in Catalonia.

8. Bibliography

The referencing style used is Vancouver, which is commonly used in the fields of medicine and science. The template for this citation method in medical journals is the following: Surname First/Middle name initial. Article title: subtitle. Abbreviated journal name date;#volume(#issue):#first page of article-#last page of article.

The template for this citation method in web-based medical journals is the following: Surname First/Middle name initial. Article title: subtitle. Abbreviated journal name [web-based journal]. Publication date [cited citation date];#volume(#issue):[page length]. Available at: URL address.

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