

*Ich habe kein besondere Begabung,  
Ich bin nur leidenschaftlich neugierig.*

*(Einstein)*



# 1 | General introduction



## INTRODUCTION

Bacterial meningitis (BM) is a severe and often life threatening infectious disease of the central nervous system (CNS) accounting for an estimated annual 170,000 deaths worldwide. It mostly affects people at the extremes of the age spectrum, infants and children on the one hand, and elderly on the other hand. The case fatality rate is estimated between 4 and 10% [1].

The etiology of BM is driven by the age of the affected patient. In neonates the most common etiologic pathogens are Group B *Streptococcus*, *Listeria monocytogenes*, *Escherichia coli*, and other Gram-negatives [1, 2]. In infants and young children worldwide, *Streptococcus pneumoniae* (SP), *Neisseria meningitidis* (NM), and *Haemophilus influenzae* type b (Hib) are the most common causes of BM. Among children older than 5 years of age and adolescents, SP and NM are the predominant causes of BM [1].

The epidemiology of BM has changed drastically over the last decades. Before routine childhood immunizations against Hib were introduced in the late 1980's and early 1990's, BM was primarily a disease of young children accounting for more than half of all BM cases [1, 3, 4]. Currently, the epidemiology of pneumococcal meningitis (PM) is changing due to the introduction of conjugate vaccines against SP in most developed countries. Besides protection of young children, it also provides herd immunity in adults, although immunity wanes over time so the age distribution of BM is momentarily shifting to the older age groups [5]. Introduction of the serogroup C meningococcal conjugate vaccine in infancy has reduced disease caused by this specific serotype, but did not influence the incidence of BM caused by the serogroups A, B, Y and W-135 [1].

The highest risk of BM caused by SP is in infants younger than 2 years old and has an incidence of approximately 20 per 100,000 [1]. The incidence of meningococcal meningitis (MM) is greatest in infants younger than 1 year old; a second peak incidence is observed at age 15 to 17 years [6]. Incidence rates vary between 1 and 2 per 100,000. In sub-Saharan Africa, meningococcus serogroup A is a major cause of meningitis epidemics [7].

In developing countries, *Mycobacterium tuberculosis* (M. tub) is a common cause for BM besides the aforementioned pathogens. In South Africa, where tuberculosis (TB) is endemic with 998 cases per 100,000, the incidence of tuberculous meningitis (TBM) ranges from 31.5 per 100,000 in children under 1 year of age to 0.7 per 100,000 in 10-14 year-olds [8].

This thesis consists of two parts and describes studies on the innate immune response in (myco) bacterial meningitis. The first part of this thesis focuses on TBM and describes a large retrospective cohort of children with TBM in South Africa, reviews

the available animal models to study TBM and presents a new animal model to study the immune response in TBM. The second part focuses on BM caused by either NM or SP and reviews the role of genetic variability in immune response genes in BM pathogenesis. It also presents several associations of polymorphisms in innate immune response genes with susceptibility to or severity of BM.

## CLINICAL PRESENTATION OF BACTERIAL MENINGITIS

Infection of the meninges and cerebrospinal fluid (CSF) is called meningitis. Clinically, meningitis presents as a severe febrile illness with signs of CNS involvement. Adults and older children often complain of severe headache and vomiting and present with a stiff neck, representing meningeal irritation. Infants usually present with inconsolable crying and irritability upon handling and often have a bulging fontanel. Both may show signs of altered consciousness.

## HISTORY

These clinical signs are recognized as a disease entity since centuries. The first publications date from 1806 by the Swiss physician Viesseux who described a meningitis outbreak in the Geneva area in 1805 [9]. In 1887 an Austrian pathologist and bacteriologist called Anton Weichselbaum was the first to isolate the causative agent of cerebrospinal meningitis, which he called *Diplococcus intracellularis meningitidis* [10]. Later the pathogen was renamed *Neisseria meningitidis* because it was his German colleague Albert Neisser who discovered *Neisseria gonorrhoeae* in 1879, which retrospectively belonged to the same family. A trivial and amusing fact given the background of this thesis, is that Neisser also co-discovered the pathogen *Mycobacterium leprae*.

## PATHOGENESIS AND TREATMENT

In the period that pathogenesis was unclear, treatment of meningitis consisted of bed rest, excessive sweating and the application of spiritual liquids. In the tradition of phlebotomy, lumbar puncture was introduced in 1891. The American pharmacist and pathologist Simon Flexner introduced meningococcal antiserum in 1913, which he applied intrathecally [11]. This therapy reduced the previous mortality of 100% to 31%. With increasing knowledge of disease causing pathogens but mostly by the

invention of antimicrobial drugs in the late 1930's, antibiotic therapy became the cornerstone of meningitis therapy and still is today.

However, with increasing knowledge of the details of pathogenesis and the inflammatory response with its intriguing ambiguous role of both microbial clearance and tissue damage, immunomodulation became the new horizon in meningitis research. The first proper study on the use of corticosteroids in meningitis was reported in 1988 [12]. Until today a lot of debate is going on the role of corticosteroids. A recent meta-analysis by the Cochrane collaboration concluded that corticosteroids significantly reduced hearing loss and neurological sequelae, but did not reduce overall mortality. Data support the use of corticosteroids in patients with BM in high-income countries. They found no beneficial effect in low-income countries [13]. A Cochrane meta-analysis of studies in TBM concluded that corticosteroids should be routinely used in HIV-negative people with TBM to reduce death and disabling residual neurological deficit amongst survivors. However, there is not enough evidence to support or refute a similar conclusion for those who are HIV positive [14].

Recently, Koedel *et al.* reviewed the current knowledge on pathogenesis of BM and focused on promising targeted approaches for adjunctive therapy, including limiting the release of toxic bacterial products (e.g. killing bacteria softly with non-bacteriolytic antibiotics) and interfering in the generation of host-derived cytotoxins by inducing neutrophilic apoptosis [15].

## HOST GENETICS OF BACTERIAL MENINGITIS

The last decade, research on susceptibility and course of infectious diseases has shifted from an environmental and microbial point of view to the host and the identification of specific genes linked to severity phenotypes of disease [16]. The next challenge will be to bring this knowledge from the proverbial laboratory bench to patient bedside. Data from host-pathogen genomic studies and molecular epidemiologic genome-wide association studies should be translated into clinical interventions and prevention programs, preferentially targeted to specific individuals at risk, based on their genetic profile (personalized healthcare). The field of Public Health Genomics is rapidly evolving and embraces the Bellagio statement (2005) which core aim is "the responsible and effective translation of genome-based knowledge and technologies into public policy and health services for the benefit of population health". In the last years this field has progressed enormously.

## AIMS AND OUTLINE OF THIS THESIS

The focus of this thesis is innate immune responses in (myco) bacterial meningitis.

Part one consists of studies on TBM. First, a retrospective cohort study of 554 children with TBM in the Western Cape of South Africa is presented. Next, a review summarizes animal models to study TBM. Furthermore, an experimental study describing a murine model to study the pathogenesis of TBM is presented.

Part two consists of a review summarizing studies on genetic variation of innate immune response genes in invasive pneumococcal and meningococcal disease applied to the pathogenesis of meningitis. Next, four studies will be described focusing on the role of single nucleotide polymorphisms (SNPs) in a cohort of 472 survivors of BM caused by either NM or SP. The first study describes a SNP in the Toll-like receptor 9 gene (*TLR9*) affecting susceptibility to develop meningitis upon acquisition of NM. A second study focuses on *TLR9* SNPs determining disease severity and the local inflammatory response inside the CNS in survivors of MM. Next, a study on BM susceptibility describes a set of SNPs in immune response genes for its respective potential to influence the development of meningitis upon infection with either meningococci or pneumococci. The last study summarizes the relation of SNPs in a set of immune response genes for its relation with disease severity in BM survivors, especially hearing loss.

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