

Lipid A variants in meningococcal disease and carriage isolates; prevalence and clinical impact

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Introduction

Neisseria meningitidis is worldwide an important cause of invasive bacterial infections. The most important trigger of the pro-inflammatory cascade accompanying meningococcal disease is the lipid A component of lipopolysaccharide. Recently, naturally occurring meningococcal mutants with under-acylated lipid A due to an inactive *lpxL1* gene have been described in adult patients with meningococcal meningitis¹. Infection with these mutant lipid A strains caused less coagulopathy, presumably due to reduced activation of Toll-like receptor 4 (TLR4).

Aim

The aim of this study was to explore the prevalence of lipid A variants among meningococcal disease and carriage isolates and to study the clinical impact of these mutants in all age-groups.

Methods

Data collection: In a retrospective cohort study of patients with invasive meningococcal diseases in the Netherlands 448 isolates were screened for lipid A mutations. All cases with invasive meningococcal disease reported by nine sentinel laboratories between June 2001 and May 2006 were included, covering circa 25% of the Dutch population. In addition, 822 carrier isolates were included from meningococcal carrier study conducted in Bavaria, Germany².

Laboratory methods: To identify meningococcal isolates as lipid A variants, isolates were tested for their capacity to induce the pro-inflammatory cytokine IL-6. Serial dilutions of heat-inactivated bacterial suspensions were added to the human monocytic cell line Mono Mac 6 (MM6) and incubated. IL-6 in supernatant was determined by ELISA. Of all strains causing a low induction of IL-6, the *lpxL1*-gene was sequenced to define the mutation.

Results

Figure 1: Of the 448 disease isolates, 29 (6.5%) had a decreased potential to induce IL-6. Median age of lipid A variants 19.3 years (IQR 4.6-50.5) compared to 5.9 years (IQR 1.8-18.9) for wild type meningococci ($p=0.007$). Proportion of lipid A variants varies from 4.4% among children under 5 years of age to 24.2% among patients 45-64 years of age

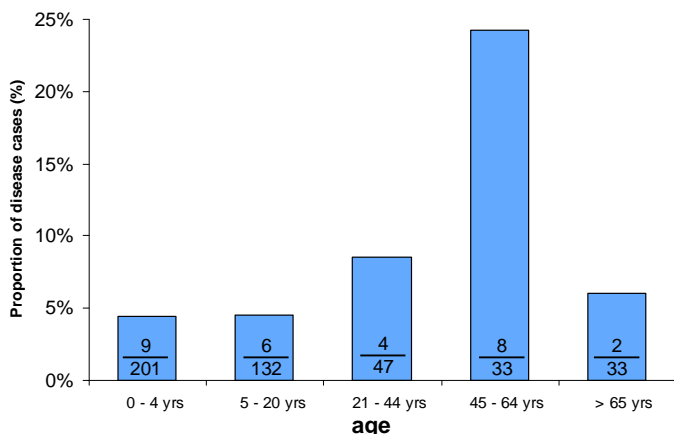


Figure 2: In patients over 5 years of age infections with lipid A variants were associated with increased underlying comorbidities, less fever and petechiae, less septic shock and decreased IC admittance compared to wild-type strains. In children under 5 years of age no clinical differences were observed between mutant and wild-type isolates.

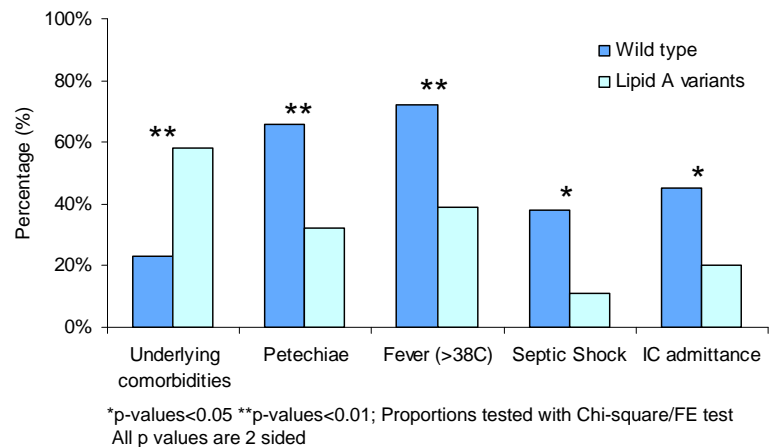
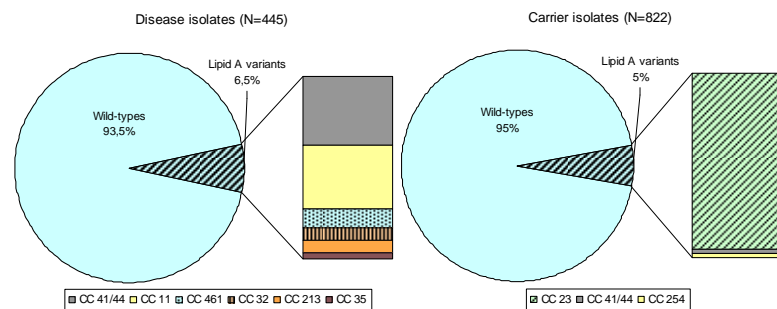


Figure 3: Among carrier isolates, 45 lipid A variants were found (5.5%). Forty-three were found among 71 isolates (61%) of clonal complex cc23. This clonal complex was not observed among disease strains.



Conclusions

- Meningococci with lipid A mutations were more frequently found in children over 5 years and adults and associated in these age-groups with increased underlying comorbidities and less severe disease.
- Among carriage isolates lipid A mutations occur only very rarely, with the exception of one single clonal complex.
- These findings shed new light on the adaptation of meningococci when interacting with its host. It emphasizes further study of lipid A variants in invasive disease and carriage, as such immune evasion may have implications for effectiveness of vaccination against meningococcal disease.

¹ Fransen et al; PLoS Pathog 2009; 5(4) ² Claus et al; J Infect Dis 2005; 191(8):1263-1271.

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