Meningococcal Disease (MD):
Epidemiology, Prevention and Diagnosis

Shao Zhujun, MD, PhD
Institute for Communicable Disease Control and Prevention, China CDC
2020-10-21
Overview

- Incidence/epidemiology of MD
- Prevention of MD: Vaccines and vaccination
- Diagnosis
Neisseria meningitidis/meningococcus

- Aerobic, gram-negative, non-motile, diplococcus
- Can cause **Meningitis** or **Septicemia** or both
  (pneumonia, arthritis, carditis)
- 12 serogroups: 6 are most virulent (A, B, C, W, X, Y)
- High morbidity and mortality
  - Up to 15% mortality even with treatment,
  - 10-15% of survivors with serious sequelae
- Incubation period 1-4 days

*N. meningitidis*
Genome size: ~2.27 Mb
Genes: ~2114
G+C: ~51.5%
Transmission of *N. meningitidis*

Risk factors for Meningococcal Disease

• *N. meningitidis* virulence factors (capsule, adhesions)

• Individual risk factors
  • Age-related acquisition of bactericidal antibodies
  • Underlying immune defects (i.e., asplenia, genetic)

• Population risk factors
  • Crowding: bar, dormitories, Household exposure
  • Smoke exposure: Active and passive smoking
  • Upper respiratory tract infections, influenza infection
  • Demographic and socio-economic factors
Invasive meningococcal disease

Meningitis

Nasopharyngeal colonization (10%)

Meningococcemia

Arthritis

Purpura fulminans

Invasive pneumopathies

pericarditis
How serious are MDs?

Sequelae: amputation, dermatorrhagia, skin lesion/scar, long term nervous system damages
MD cases in China
Typing of *Neisseria meningitidis*

**Phenotyping**
Capsule (12 serogroups)
12 serogroups, 6 were known to cause epidemics (A, B, C, W, Y, X)

**Genotyping**
- MLST: polymorphism of several genes.
- Whole Genome sequencing (WGS)
Epidemiology of Meningococcal disease
<table>
<thead>
<tr>
<th>serogroups</th>
<th>clonal complex (cc)</th>
<th>epidemic</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>cc 1, cc 5</td>
<td>2 epidemics</td>
</tr>
<tr>
<td>B</td>
<td>cc 8, cc 11, cc 32, cc 41/44</td>
<td>Europe American Australia</td>
</tr>
<tr>
<td>C</td>
<td>cc 8, cc 11, cc 32, cc 41/44</td>
<td>Europe American Australia</td>
</tr>
<tr>
<td></td>
<td>cc 4821</td>
<td></td>
</tr>
<tr>
<td>W135</td>
<td>cc 11</td>
<td>2000, Hajj outbreaks</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2006, China</td>
</tr>
<tr>
<td>Y</td>
<td>cc 23, cc 167</td>
<td>United American</td>
</tr>
<tr>
<td>X</td>
<td>cc 181</td>
<td>African outbreaks</td>
</tr>
</tbody>
</table>
The “Meningitis Belt” of sub-Saharan Africa

• African meningitis belt

✔ 80 % of the global burden : > 1.3 million cases 1986-2015
✔ Seasonal hyper-endemic activity
✔ Cyclical large-scale epidemics, reaching annual incidence rates > 100 cases per 100,000 population.
✔ Control program tailored for this area

millions are at risk

*Meningitis belt* defined by Lapeyssonnie in 1963
Redefined by Greenwood in 1987
Meningitis Epidemics in African Meningitis Belt

- *Neisseria meningitidis*
  - Several epidemic-causing serogroups (A, C, W, X)
- **NmA** responsible for majority (>80%) of epidemics in the African meningitis belt until 2010.
- **Epidemics** also due to C, W and X
Epidemiology of MD in China

- 1950s: A, ST-5
- 1960s: A, ST-3
- 1960s: A, ST-7
- 1980s: C, ST-4821
- 2003: W, ST-11
- 2006: B, ST4821
- 2015: Y, X
- ?
Prevention of MD--vaccine and vaccination
Meningococcal Vaccines

- **Polysaccharide Vaccine**
  - Protects against serogroups A, C, Y, W (not serogroup B)
  - Poorly immunogenic in children <2 yrs
  - Immunity of limited duration <3 yrs

- **Conjugate Vaccine**
  - Protects against serogroups A, C, Y, W (not serogroup B)
  - Better immunogenic in children <2 yrs
  - Immunity of limited duration ≥3 yrs

- **MenB vaccine**
The Meningitis Vaccine Project and MenAfriVac® immunization programme in the African meningitis belt
MenAfriVac introduction strategy

• **Inducing strong herd protection**
  - Large single dose mass vaccination campaigns with high coverage, targeting 1–29 year-olds in 26 belt countries

• **Protecting new birth cohorts**
  - Routine EPI immunization or periodic follow-up campaigns

• **Enhancing surveillance and outbreak response capacity**
  - Throughout vaccine introduction + beyond
  - Rapid response to outbreaks (W, C, X and A in the unprotected)
  - Adequate care/treatment of meningitis cases
  - Outbreak containment: emergency stockpile
Official launch day – health workers
Official launch day – school children
MenAfriVac rollout 2010–2016

Years of Vaccination Campaign:
- 2010 Phase 1
- 2010 Phase 2
- 2011
- 2012
- 2013
- 2014
- 2015
- 2016

Bar chart showing the increase in millions of persons vaccinated from 2010 to 2016:
- 2010: 19
- 2011: 55
- 2012: 103
- 2013: 154
- 2014: 217
- 2015: 237
- 2016: 277
Meningitis pathogen trends in the belt, 2006-2015

MenAfriVac introduction

Courtesy C. Lingani, WHO/AFRO IST-West
MenAfriVac campaigns, A huge success

- **Excellent coverage**
  - High population acceptance
  - Reported coverage usually >95%
  - Age group > 15 years often less well covered
  - Mop-up campaigns conducted in areas with lower coverage (Senegal, Cameroon)

- No new serious AEFI attributable to the vaccine detected
- Vaccine wastage < 5%
- Operational costs **0.65 USD / target person**
WHO recommendations

• Countries completing mass vaccination campaigns introduce meningococcal A conjugate vaccine into the routine childhood immunization programme within 1-5 years following campaign MenAfriVac 5 μg should be used for routine immunization of infants and young children from 3 to 24 months of age.

• A one-time catch-up campaign should be conducted for birth cohorts born since the initial mass vaccination and outside the age range targeted by the routine immunization programme MenAfriVac (10 μg) should continue to be used for catch-up and periodic campaigns from 12 months of age onwards.
Surveillance and Diagnosis
Objective of MD surveillance

- Outbreak detection for response
- Describe the epidemiology of meningitis
  - Incidence trends
  - Monitor the circulation and distribution of Nm serogroups and strains (sequence-type)
  - Monitor antibiotic susceptibility
- Provide data to estimate disease burden
- Identify geographical areas and populations at risk
- Allow measuring vaccine effectiveness
- Estimate the impact of preventive vaccination on the disease
  - Impact on serogroup circulation
Surveillance essential

- **Epidemiology**: surveillance systems
  - Adapted to the objective: countrywide or sentinel
  - Implementation depends on disease burden

- **Laboratory**
  - Transportation of specimen
  - Rapid confirmation of pathogen is critical to determine appropriate treatment and epidemic response.

- **Linking of epidemiological and laboratory data**
Laboratory Testing for Vaccine Preventable Diseases

- **As disease incidence declines**
  - Clinical diagnosis less specific
  - Laboratory confirmation more important
    - Critical to determine serogroups/types or genotypes

- **Accurate diagnosis required to**
  - Develop best vaccination policy
  - Measure vaccine impact
  - Measure vaccine effectiveness
  - Monitor changes in serogroups/types (replacement or emergent strains) and antibiotic resistance
  - Refine vaccination strategy
WHO- Global IB-VPD Surveillance Network

- **Invasive Bacterial Vaccine-Preventable Disease (IB-VPD)**
  - **Global sentinel** site case-based surveillance network
  - Meningitis (pneumococcus, Hib, and meningococcus)
  - Initially mainly established to monitor impact of Hib and PCV vaccination program
  - Pediatric hospitals only (children up to 5 years)
  - Coordinate clinical, laboratory and data management
- Coordinated by WHO with partners since 2008 with Gavi funding
Case definition

Confirmed case

- Isolation of *N. meningitidis* from a normally sterile site (e.g., **blood or cerebrospinal fluid [CSF]**) from a person with clinically compatible illness.
- Detection of *N. meningitidis* DNA by PCR or Polysaccharide antigen in CSF (e.g., by latex agglutination)
Diagnosis: clinical specimens

- Lumbar puncture (Cerebrospinal fluid sample, CSF)
- Blood
- Sputum
- Swabs
CSF specimens

Process of CSF

Transport to laboratory < 1 hour

Centrifuge at 1000 x g for 10 to 15 minutes

CSF Specimen

Freeze 250 µl at -70°C for rt-PCR

Supernatant

Transport to laboratory > 1 hour

Inoculate Trans-Isolate (TI) medium

Incubate overnight 35°C in CO₂

Sediment

Subculture to chocolate agar and blood agar

Primary plating on chocolate and blood agar

Rapid diagnostic test (RDT)

Gram stain

Latex agglutination
Distribution of CSF at hospital (with T-I protocol)

Collect at least 3.0 mL CSF

**First priority**
- Transfer 1.5 mL to a tube
- Transfer 0.75 mL to T-I vial. *Keep at room temp.*

**Second priority**
- Transfer rest to a tube. *Keep at 4-8°C*

**Third priority**
- In-hospital diagnostic tests
- Transport to provincial CDC lab. *Keep at room temp.*
- Transport to prefecture CDC lab. *Keep at 4-8°C*

---

1 Appropriately labeled.
2 Tests done in hospital lab: Protein/glucose (0.15 mL); Cell count (0.25 mL); Bacteriology (0.5 mL): Gram stain, latex agglutination, culture
3 Tests done at provincial CDC lab: Bacterial culture
4 Tests done at prefecture CDC lab: JE IgM ELISA (0.1 mL) and storage (the remaining CSF)
Distribution of blood at hospital

Collect into each of two tubes without anticoagulant:
- 5-10 ml from adult
- 2-5 ml children
- 0.5-2 ml infants

First tube

- Inoculate 3 blood culture bottles. *Keep at room temp.*

Second tube

- Separate serum and transfer to tube1. *Keep at 4-8°C*

In-hospital bacterial culture

Transport to prefecture CDC lab *Keep at 4-8°C*

---

1 Appropriately labeled.
2 Tests done at in-hospital lab: Bacterial culture
3 Tests done at prefecture CDC lab: JE IgM ELISA (0.1 mL) and storage (the remainder)
Primary Plating

Blood agar plate

Chocolate agar plate
Identification of *Neisseria meningitidis*

Carbohydrate utilization test

<table>
<thead>
<tr>
<th>Organism</th>
<th>Glucose</th>
<th>Maltose</th>
<th>Lactose</th>
<th>Sucrose</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Neisseria meningitidis</em></td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>Neisseria lactamica</em></td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Neisseria gonorrhoeae</em></td>
<td>+^2</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>Neisseria sicca</em></td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td><em>Moraxella catarrhalis</em></td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>
Slide agglutination serogrouping
Storage of *N. meningitidis* strains

**Temporary**
- Silica gel packages
- TI medium
- Chocolate agar slants

**Permanent**
- Freezing isolates in defibrinated sheep or rabbit blood at -70°C
- Lyophilization
### PCR diagnosis

<table>
<thead>
<tr>
<th>Serogroup</th>
<th>Capsule type</th>
<th>Gene Target Name</th>
<th>Alternate Gene Names</th>
<th>Ref</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>(α1→6)-N-acetyl-D-mannosamine-1-phosphate</td>
<td>sacB</td>
<td>staD, staD of B, siaDg</td>
<td>(31)</td>
</tr>
<tr>
<td>B</td>
<td>(α2→8)- N-acetyleneuraminic acid</td>
<td>synD</td>
<td>siaD of C, siaDc</td>
<td>(5, 9, 50, 58, 61)</td>
</tr>
<tr>
<td>C</td>
<td>(α2→9)- N-acetyleneuraminic acid</td>
<td>synE</td>
<td>siaD of W135, siaDπ</td>
<td>(4, 9, 14, 35, 49, 61)</td>
</tr>
<tr>
<td>W135</td>
<td>6-D-Gal(α1→4)-N-acetyleneuraminic acid(α2→6)</td>
<td>synG</td>
<td>siaD of W135, siaDπ</td>
<td>(4, 9, 14, 35, 49, 61)</td>
</tr>
<tr>
<td>X</td>
<td>(α1→4)-N-acetyl-D-glucosamine-1-phosphate</td>
<td>xcbB</td>
<td>(2, 6)</td>
<td></td>
</tr>
<tr>
<td>Y</td>
<td>6-D-Glc(α1→4)-N-acetyleneuraminic acid(α2→6)</td>
<td>synF</td>
<td>siaD of Y, siaDy</td>
<td>(4, 9, 14, 35, 49, 61)</td>
</tr>
</tbody>
</table>

- **CSF, blood, serum, etc**
- **Isolate or Trans-Isolate**
- **Prepare DNA Template**
- **Perform species-specific real-time PCR assays: sodC or ctrA, hpd, and lytA**
  - If *sodC* (or *ctrA*) positive: Perform serogroup-specific real-time PCR assays for serogroups A, B, C, W135, X and Y.\(^1\)
  - If *hpd* positive: Perform serotype-specific real-time PCR assays for serotypes a, b, c, d, e, f.\(^2\)
  - If *lytA* positive: Perform conventional multiplex PCR assays to determine serotype. Use Tables 6-8 to choose scheme based on geographical region.\(^3\)
MLST: Neisseria meningitidis

This site uses two linked databases powered by the BIGSdb genomics platform. The sequence definition database contains allele sequence and MLST profile definitions whereas the isolate database contains provenance and epidemiological information. Further details about BIGSdb can be found in Jolley & Maiden 2010, BMC Bioinformatics 11:1595.

As well as MLST, the platform contains sequence data for other genetic targets, allowing MLST, PorA, porB, FetA, rfb, penA, rpoB and other sequences to be queried from the same interface.
Whole Genome sequencing (WGS)

Reinaldo Acevedo, 2018, Expert Review of Vaccines
Summary

• Epidemiology of disease varies by region
• Case fatality ratio high globally
• Burden and impact of disease in Africa “Meningitis Belt” substantial
• Analysis limited by lack of laboratory-based serogroup surveillance
Acknowledgements

Adapted from:

- Ray Borrow, UK
- Varja Grabovac, WPR
- Leonard W. Mayer, USCDC
- Dr. Stephen C. Hadler, USCDC
- Olivier RONVEAUX,
- AMES partners
- GMI partners