Epidemiology of Hand, Foot and Mouth Disease in South-East Asia & Key Immunological Considerations for Vaccine Development

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31 October - 1 November 2023, WHO/MPP mRNA regional workshop in Bangkok, Thailand
Hand, Foot and Mouth Disease (HFMD)

General
- Highly contagious
- Common in young children
- Group of enteroviruses - coxsackievirus A viruses, enterovirus A71, echoviruses
- Pathogenesis
  - faecal-oral, direct
  - replicate in oropharynx
  - viraemia and dissemination to target organs (CNS, skin)
  - excreted in pharynx and faeces for weeks

Symptoms
- Fever, sore throat, mouth ulcers
- Herpangina vs HFMD
- Blisters on palms of hands and soles of feet
- Symptoms usually appear 3 to 5 days after exposure
- Recurrent HFMD - 0.45%\(^4\)

Complications
- Rare neurological complications
- Aseptic meningitis, brain stem encephalitis with neurogenic edema
- In infants and young children (mean age < 2 years old)
- More commonly associated with EV-A71 (0.1-1.1% severe; 0.01-0.03% fatal)\(^1, 2\)
- Long-term neurological sequelae\(^3\)

References:
Enteroviruses

- Family of *Picornaviridae*
- Genus *Enterovirus*
- Single-stranded positive sense RNA (~7.4 kb)
- Capsid proteins VP1 – VP4
- VP1-3 receptor binding, antigenicity
- Non-structural polyprotein processing, replication
- Receptors- SCARB2, PSGL-1, heparan sulfate etc
Epidemiology & Immunological Considerations

- Disease burden of HFMD
- Known target population
- Lessons from polio and current EV-A71 vaccines
- Neutralizing antibody correlate of protection
- Durability of immune protection
- Immunogenicity requires complete capsid & is species specific
Disease Burden of HFMD

China: HFMD morbidity cases remained at approximately 2 million

Malaysia: Second most common infectious disease

- Total of 94,313 hospitalized HFMD cases
- HFMD economic burden- US$90,761,749


Ministry of Health Malaysia.

Open Forum Infectious Diseases 2019; 6: ofz284
HFMD affects young children

Reported HFMD cases in Malaysia

Age vs. Year

Number of HFMD cases:
- 0
- 10,000
- 20,000
- 30,000
- 35,043

Personal communication, MOH Malaysia
## EV-A71 Vaccines- Good Neutralizing Antibodies & Durable Immune Protection

<table>
<thead>
<tr>
<th>Organizations</th>
<th>Cell Line</th>
<th>Strain</th>
<th>Dosage (mg)</th>
<th>Population Target In trial</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sinovac Biotech Co., Ltd (China)</td>
<td>Vero cell</td>
<td>C4a (H07 strain)</td>
<td>1</td>
<td>6–35 months old</td>
<td>94.8% efficacy Approved Dec 2015</td>
</tr>
<tr>
<td>Beijing Vigoo Biological Co., Ltd. (China)</td>
<td>Vero cell</td>
<td>C4a (FY7VP5 strain)</td>
<td>0.8</td>
<td>6–35 months old</td>
<td>97% efficacy Approved Dec 2016</td>
</tr>
<tr>
<td>CAMS (China)</td>
<td>KMB- 17 cell</td>
<td>C4a (FY- 23 K-B strain)</td>
<td>0.25</td>
<td>6–71 months old</td>
<td>94.7% efficacy Approved Dec 2015</td>
</tr>
<tr>
<td>NHRI (Taiwan)</td>
<td>Vero cell</td>
<td>B4</td>
<td>5 and 10</td>
<td>20–43 years adults</td>
<td>-</td>
</tr>
<tr>
<td>Enimmune Co. (Taiwan)</td>
<td>Vero cell</td>
<td>B4</td>
<td>0.25, 0.5, 1, 2 and 5</td>
<td>6 month –6 years old</td>
<td>-</td>
</tr>
<tr>
<td>Medigen Vaccine Biologics Co. (Taiwan)</td>
<td>Vero cell</td>
<td>B4</td>
<td>150</td>
<td>2 month - 6 years old</td>
<td>96.8% efficacy Approved 2023</td>
</tr>
<tr>
<td>Inviragen (Singapore)</td>
<td>Vero cell</td>
<td>B4</td>
<td>0.6 and 3</td>
<td>Adults</td>
<td>Terminated</td>
</tr>
</tbody>
</table>

Immune Responses (EV-A71)

**Innate**
- Cytokine storm associated with IL-1 beta, IL-6, IL-10, IL-17A, MCP-1, IL-8 MIG, IP-10, G-CSF
- **activated by non-structural viral proteins**

**Humoral**
- **Maternal antibodies** decline by 6 months
- IgM-detectable 3-6 days after onset, **cross-reactive**
- 1:16-1:32 neutralizing antibody as immunological surrogate endpoint for EV71 vaccine protection
- Seropositivity and seroconversion after two vaccine doses were ~100% , >IU 36.2 IU/ml
- IVIG for treatment
- **dominant linear epitopes in mice and human are different**
- antibody-dependent enhancement only in vitro model

**Cellular**
- Correlates with disease progression and clinical outcome
- Th1 & pro-inflammatory cytokines in blood
  - **Cross-reactive epitopes**

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2. Human Vaccine and Therapeutics 2022, 18: e2073751.
• Antigenic- **F-antigen** (D antigen in poliovirus)
• **Antigenic loops** - insertion, replacement
• The structural protein **VP1** is the main immunodominant site targeted by anti-EV-A71 IgM and IgG antibodies
• **DE loop**- EV-A71 IgM-specific immunodominant epitope
• Epitopes against **non-structural proteins** were also detected
• Antisera from **rabbits** injected with formalin-inactivated EV-A71 only recognized VP2-28 peptide (aa 136–150 VP2)
• **Mouse** antisera only recognized VP1-43 (aa 211–225 VP1= SP70)
- different but good/acceptable cross-reactivity between genogroups
- species-specific dominant epitopes
- careful interpretation of animal studies

Ev-A71 Antigenicity, Immunogenicity & Immune Protection

Mouse

Rabbit

Human

Antiviral research 2023, 212: 10559
Vaccine 2009, 27: 3153–3158
PLoS Negl Trop Dis 2014, 8: e3044
Epidemiology & Immunological Considerations

- Disease burden of other enteroviruses
- Antigenic diversity, Multiple serotypes and genotypes
- Virulence & Recombination
- Multivalent candidates
- Immune Interference
HFMD in Asia- Presence of Multiple Enteroviruses- The Need for Multivalent HFMD Vaccine

Modified from Pediatric Infectious Disease Journal (2016) 35: 285- 300
Virologica Sinica 2020, 35: 21-33, Biosafety and Health 2019, 1: 32-40
Personal communication with National Public Health Laboratory, Malaysia
## Characteristics of HFMD in Mainland China: Before and After Vaccine

<table>
<thead>
<tr>
<th></th>
<th>2013-2015 (Before)</th>
<th>2017-2019 (After)</th>
<th>Change (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Incidence rates *</td>
<td>2800.78</td>
<td>2575.29</td>
<td>-8.05</td>
</tr>
<tr>
<td>Severe illness rates *</td>
<td>18.97</td>
<td>7.17</td>
<td>-62.20</td>
</tr>
<tr>
<td>Mortality rates *</td>
<td>0.37</td>
<td>0.06</td>
<td>-83.78</td>
</tr>
<tr>
<td>Severe/Cases (%)</td>
<td>0.68</td>
<td>0.28</td>
<td>-58.82</td>
</tr>
<tr>
<td>Death/Cases (%)</td>
<td>0.01</td>
<td>0.00</td>
<td>-100.00</td>
</tr>
<tr>
<td>Death/Severe Cases (%)</td>
<td>1.97</td>
<td>0.85</td>
<td>-56.85</td>
</tr>
</tbody>
</table>

* * indicates significance at p < 0.05

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**First inactivated EV-A71 vaccine was approved**

- **At the end of 2015**
  - 62.80% EV-A71
  - 37.20% Non-EV-A71
- **From 2013 to 2015**
  - 32.80%
- **From 2017 to 2019**
  - 67.20%

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**Laboratory Results**

- CV-A10
- EV-A71
- Other enterovirus

**Graphs:**

- **A** All
- **B** Mild
- **C** Severe

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The Lancet Regional Health - Western Pacific 2022;20: 100370, Vaccine 2021, 39: 3319–3323
Circulation of Other Enteroviruses-USA

USA Enterovirus Surveillance (by year)

Do we need to add other vaccine candidates?

Data from CDC https://www.cdc.gov/non-polio-enterovirus/resources.html
Monitoring virulence & recombination in EV-A71

- Severe HFMD Vietnam- C4\(^1\), less severe Malaysia- B4\(^2\)
- Virulence in SCARB2-transgenic mice clade B5 (I, II), C4 (II-IV)\(^3\)
- Virulent mutations VP1 145, 244\(^4\)
- Recombination alters fitness and virulence\(^5\)

1. Eurosurveillance 2018, 23: 1800590
2. Clinical Infectious Diseases 2007; 44: 646-656
Learn from Multivalent HFMD Vaccines

**TYPES**

- Inactivated
- Live-attenuated
- Subunit-peptide, VP1 etc
- Virus-like particles
- Bivalent/Trivalent/
  Tetravalent/
  Hexavalent

**CHARACTERISTICS**

- HFMD disease burden: EV-A71, CV-A6, CV-A10, CV-A16
- HFMD and other enteroviruses: CV-B, Echo
- Innate and cellular responses- non-structural viral proteins
- Cross protection- conserved regions
- Balance Immune interference

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*Therapeutic Advances in Vaccines and Immunotherapy* 2019;7.
*Hum Vaccin Immunother* 2015, 11:2688-704
*Vaccines* 2014, 32: 6177-6182.
Infection with EV-A71 or CV-A16 serotype provide transient immunity against each other.

Vaccination - Serotype replacement by CV-A16 transient, big reduction in the burden of EV-A71-associated HFMD.

A mass EV-A71 vaccination program of infants and young children will reduce HFMD burden.

Possible asymmetry in its strength such that CV-A16 serves as a stronger forcing on EV-A71.
Summary: Epidemiology & Immunological Considerations

**STRENGTHS**

- Disease burden of HFMD
- Known target population
- Lessons from polio and current EV-A71 vaccines
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- Durability of immune protection
- Immunogenicity requires complete capsid & is species specific

**FUTURE IMPROVEMENTS**

- Disease burden of other enteroviruses
- Antigenic diversity, Multiple serotypes and genotypes
- Virulence & Recombination
- Multivalent
- Immune Interference