History and Evolution of Vaccine Technologies

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Immunize Nebraska

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Disclosure

Employed by GSK where I am a vaccine research physician scientist

Presentation at the invitation of Dr. Meera Varman, Professor, Pediatric Infectious Diseases, Creighton University School of Medicine

Presentation is for educational purposes only; this is not a sales, marketing or promotional presentation

Content of presentation will not include unapproved or investigational uses of products or devices
Globally, Many Diseases are Currently Preventable by Vaccination

“Vaccines are one of the greatest achievements of biomedical science and public health”

<table>
<thead>
<tr>
<th>Global public health¹</th>
<th>Regional focus²</th>
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</thead>
<tbody>
<tr>
<td>Diphtheria</td>
<td>Adenovirus</td>
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<tr>
<td><em>Haemophilus influenza type b</em></td>
<td>Anthrax</td>
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<tr>
<td>Hepatitis A</td>
<td>Cholera</td>
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<tr>
<td>Hepatitis B</td>
<td>Dengue</td>
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<td>Herpes zoster</td>
<td>Ebola</td>
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<tr>
<td>Human papillomavirus</td>
<td>Japanese encephalitis</td>
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<tr>
<td>Influenza</td>
<td>Tick-borne encephalitis</td>
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<tr>
<td>Measles</td>
<td>Typhoid fever</td>
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<tr>
<td>Meningococcal A, B, C, W, Y</td>
<td>Rabies</td>
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<tr>
<td>Mumps</td>
<td>Yellow fever</td>
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<td>H5N1 flu</td>
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<td>Pertussis</td>
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<td>Poliomyelitis</td>
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<td>Pneumococcal</td>
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<tr>
<td>Rotavirus</td>
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<td>Smallpox and vaccinia</td>
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<tr>
<td>Tetanus</td>
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<td>Tuberculosis</td>
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<td>Varicella</td>
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Vaccine Science: Two Centuries of Continuous Research, Improvements, and Achievements

The Science of Immunology Began in the 19th Century

Microorganisms were identified as the true cause of infectious diseases

Host cells that ingest and destroy invading microbes were discovered and called ‘phagocytes’

Passive immunotherapy of diphtheria with anti-diphtheria toxin antibodies discovered ‘immune serum’

The antibodies theory was formulated

The concept of ‘natural immunity’ to infection was born

New Technologies Make Vaccines Possible That Were Previously Impossible

1930
Empirical Approach
Diphtheria, Tetanus, Pertussis, Rabies, Influenza, Smallpox, Polio, BCG

1980
Recombinant DNA
Hepatitis B, Acellular Pertussis, Lyme, Human papillomavirus

1990
Glycoconjugation
MenACWY, Pneumo, Hib, GBS, S. aureus

2010
Reverse Vaccinology
MenB, GBS, GAS, E. coli, S. aureus, C. difficile

In the 1930s, Max Theiler at Rockefeller Foundation used an egg system for the development of an effective yellow fever vaccine, for which he was awarded the Nobel Prize in Medicine in 1951.

Enders, Weller and Robbins at Harvard received the Nobel Prize in Medicine in 1954 for demonstrating the ability of polio viruses to grow in cell cultures.
Virus Vaccines

Live-attenuated virus
Viruses are weakened by being passed through animal or human cells until they gain mutations that limit their ability to cause disease

Inactivated virus
Virus is inactivated using chemicals or heat

APC, antigen-presenting cell.
Live-attenuated, combined MMR vaccine developed in order to minimize the total number of injections in infants. Clinical trial data demonstrate a combined antigen vaccine can be effective and can have an acceptable safety profile.

MMR, measles-mumps-rubella
Split Pathogen and Subunit Vaccines

- Antigen choice: provides immune protection & technologically achievable
- Often reduced immunogenicity versus whole pathogen
- Non-infectious, low reactogenicity, acceptable tolerability
- No or limited availability of innate defensive triggers
- For subunit vaccines with lower immunogenicity, adjuvants often needed to compensate
- Facilitate supply via synthetic production versus whole pathogen

Whole virus vaccine

Pathogen fragmentation

Split vaccine

Subunit/purified vaccine

eg, some influenza vaccines

eg, acellular pertussis components: PT, PRN, FHA, FIM; HA in some influenza vaccines

New Technologies Make Previously Impossible Vaccines a Reality

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Recombinant Protein Vaccines: HBV

Hepatitis B surface antigen

Insert gene into yeast expression system

Purification of recombinant protein encoding antigen (natural assembly into spheres)

Administration as vaccine

HBsAg = hepatitis B surface antigen; HBV = hepatitis B virus.

Yeast DNA

Antigen DNA

DNA encoding L1

HPV viral capsid

Self-assembly into viral capsids (VLPs)

Self-assembly into pentamers

Purification from expression system

L1 capsid proteins

Translation

Transcription

Elicit immune response

1990s

Recombinant Protein Vaccines: HPV

HPV, human papillomavirus; VLP, virus-like particle

Polysaccharide-conjugate vaccines

1980s-1990s

Polysaccharide vaccine

B-cell

Antibody, no memory

Conjugated protein

Polysaccharide-conjugate vaccine

T-cell

Antibody and memory

Polysaccharide-conjugate vaccine

B-cell + antibody quantity

Polysaccharide vaccine

No T-cell response to polysaccharide alone

T-cell quantity

Classical Vaccinology

Growing Pathogens

Reverse Vaccinology: Human Immunology Instructs Vaccine Antigen Design

Classical Vaccinology

growing pathogens

Reverse Vaccinology

design from information

Reverse Vaccinology: A Genomic Approach for a Meningococcus B Vaccine\textsuperscript{1,2}

600 potential vaccine candidates identified

350 proteins successfully expressed in \textit{E. coli}

91 novel surface-exposed proteins identified

28 novel proteins have bactericidal activity

Vaccine Candidates

Vaccines Today: An Explosion of New Technologies

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2015
Next Generation Technologies
Structural Vaccinology
Adjuvants
Synthetic Biology/RNA/DNA Vaccines
Vectors
Systems Vaccinology

Current Challenges for Vaccines

<table>
<thead>
<tr>
<th>Challenging populations due to impaired immune system (e.g., elderly, children, immunocompromised)</th>
<th>Need for booster vaccinations</th>
<th>Recombinant antigens generally less immunogenic than live or attenuated organism vaccine</th>
<th>Pathogens that require broad and complex immune response</th>
<th>Need for antigen sparing potential supply problems (e.g., pandemic flu, SARS-CoV-2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increase the level of the immune response</td>
<td>Prolong the duration of the immune response, improve immune memory, and protection</td>
<td>Overcome a weakened immunogenicity</td>
<td>Induce the generation of a high and broad immune response</td>
<td>Reduce the amount of antigen needed (dose-sparing)</td>
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</tbody>
</table>

### Examples of Novel Approaches to Vaccine Design

<table>
<thead>
<tr>
<th>DNA(^1)</th>
<th>Live vectors(^1)</th>
<th>Reverse vaccinology(^1)</th>
<th>RNA(^2)</th>
<th>Novel adjuvants and adjuvant combinations(^3)</th>
</tr>
</thead>
</table>
| • Pathogen-derived genetic material coding for the antigens contained in a non-replicating DNA plasmid  
• Antigen is expressed by the cells of the vaccine recipient | • Targeted antigens encoded by gene(s) incorporated into the vector’s genetic material  
• Antigens expressed by a vector (like virus or bacterium) that is non-pathogenic | • Computer analysis of the pathogen’s entire genome is conducted to find genes that may be antigenic  
• Vaccine candidate identified based on prediction of protein sequences similar to pathogen’s genome sequences | • Synthetic virus particles include antigen proteins  
• Once inside host cell cytoplasm, these self-amplify in large amounts, express antigen proteins and interact with the host immune system | • Substances included in a vaccine formulation to enhance the quality and strength of the immune response induced by the vaccine antigen(s) |

Adjuvant$^{1,2}$

- From Latin, *adiuvare*: to aid
- Substance included in a vaccine to enhance and modulate the quality and/or strength of the immune response induced by the antigen
- Old technology, made new

Adjuvants have a long history in the fight against infectious diseases

Hib = Haemophilus influenzae type b; HPV = human papillomavirus; IPV = inactivated polio vaccine; OPV = oral polio vaccine (live).

*IPV is adjuvanted when formulated in combination with diphtheria, tetanus, pertussis-based vaccines but is not adjuvanted when formulated as a stand-alone vaccine.

Rationale for adjuvant use in vaccines
Why do antigens need help?

Illustrative figure based on Strugnell R et al. 2011; Garçon N et al. 2011

Adjuvants are associated with several potential benefits for vaccine development

- Overcome poor immunogenicity
  - e.g. Malaria Vaccine

- Increased duration of immune response
  - e.g. HPV, Herpes zoster V

- Dose reduction and antigen sparing
  - e.g. Hepatitis B V, Pandemic flu V

- Improved responses in special populations
  - e.g. Seasonal flu V, Herpes zoster V

References:
### Adjuvants in Licensed Products

<table>
<thead>
<tr>
<th>Adjuvant</th>
<th>Mechanism or Receptor</th>
<th>Licensed product</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aluminum salts</td>
<td>Nalp3, ITAM, antigen delivery</td>
<td>Numerous (eg, pertussis, hepatitis, pneumococcal)</td>
</tr>
<tr>
<td>AS04</td>
<td>TLR4</td>
<td>HPV</td>
</tr>
<tr>
<td>Emulsions (MF59, AS03)</td>
<td>Immune cell recruitment, antigen uptake</td>
<td>Influenza</td>
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<tr>
<td>AS01</td>
<td>TLR4, inflammasome</td>
<td>Zoster</td>
</tr>
<tr>
<td>CpG ODN</td>
<td>TLR9</td>
<td>Hepatitis B</td>
</tr>
</tbody>
</table>

### Adjuvants in Development

<table>
<thead>
<tr>
<th>Adjuvant</th>
<th>Mechanism or receptor</th>
<th>Clinical phase</th>
</tr>
</thead>
<tbody>
<tr>
<td>ISCOMs (Matrix-M)</td>
<td>Unknown</td>
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</tr>
<tr>
<td>dsRNA analogues</td>
<td>TLR3</td>
<td>1</td>
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<tr>
<td>Flagellin</td>
<td>TLR5</td>
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<tr>
<td>C-type lectin ligands</td>
<td>Mincle, Nalp3</td>
<td>1</td>
</tr>
<tr>
<td>CD1d ligands</td>
<td>CD1d</td>
<td>1</td>
</tr>
<tr>
<td>GLA-SE</td>
<td>TLR4</td>
<td>1</td>
</tr>
<tr>
<td>IC31</td>
<td>TLR9</td>
<td>1</td>
</tr>
<tr>
<td>CAF01</td>
<td>Mincle, antigen delivery</td>
<td>1</td>
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</tbody>
</table>

Vaccine Game-changing Technologies

Life science revolution driven by insights and data

- Adjuvant Systems
- Synthetic vaccines (DNA/RNA)
- Viral Vectors
- Reverse vaccinology
- Structural vaccinology
- Platform Technologies

Potential to improve efficacy; faster, simpler, more efficient development & manufacturing
Reverse Vaccinology and beyond

- Reverse Vaccinology was first successfully applied to MenB vaccine challenge\(^1\)

- Subsequently Reverse Vaccinology was applied to other difficult pathogens, e.g.
  - *E. coli* (Gomes Moriel D et al, PNAS, 2010)

- Antigens identified by RV or other methods can potentially be further improved using new technologies e.g. Structural Vaccinology, Reverse Vaccinology 2.0\(^2\)

Structural Vaccinology

Example - Respiratory Syncytial Virus (RSV)

- Can now use 3D knowledge of protein structure to design new vaccine antigens with optimized biological and immunological features\(^1\)
- E.g. design of RSV F antigen engineered as stable pre-fusion conformation\(^2\)

Concept of nucleic acid-based vaccines

Nucleic acid vaccine

- Express target antigens in the vaccinee
- Transformative,\(^1\) simpler technology\(^2\)
  - **No requirement** for cell culture-based manufacturing
- **Scalable** and flexible
- Types of synthetic nucleic acid-based vaccines
  - Plasmid DNA
  - messenger RNA

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Nucleic Acid Vaccines

DNA vaccine

RNA vaccine
(with lipid coat)

The DNA or RNA encoding a viral peptide that promotes an immune response is inserted into human cells, which then produce large quantities of the specific DNA or RNA.

APC, antigen-presenting cell; DNA, deoxyribonucleic acid; mRNA, messenger RNA; RNA, ribonucleic acid.
Viral vectors retain immune-stimulating properties of viruses without causing disease

Triggering the innate immune response

Early, non-specific

- Organized structure and size ideal for pattern recognition receptors ➔ efficient triggering of innate immunity\(^1\)
- Non-pathogenic & no replication in host due to removal of essential genes\(^1\)
- Infect many cell types, including Antigen Presenting Cells (APC)\(^2\)

Triggering the adaptive immune response

Long lasting, specific

- Antigen of interest inserted into viral genome\(^2\)
- Antigen produced within APC triggering antibodies and cellular immune responses\(^1,2\)

Viruses such as measles or adenovirus are weakened so they cannot cause disease, and are genetically modified to produce target antigen proteins.
Platform technologies can act as ‘on-demand’ solutions to produce vaccines against multiple diseases

Traditional vaccinology\(^1\)

- One technological development
- Multiple rounds of refinement
- One vaccine candidate

Platform-based vaccine development\(^2\)

- One technological platform
- Repurpose existing tools
- Accelerated development
- Several vaccine candidates

How Can These Technologies Help Our Society?

Empirical Approach
- Diphtheria, Tetanus, Pertussis, Rabies, Influenza, Smallpox, Polio, BCG

Recombinant DNA
- Hepatitis B, Acellular Pertussis, Lyme, Human papillomavirus

Glycoconjugation
- MenACWY, Pneumo, Hib, GBS, S. aureus

Reverse Vaccinology
- MenB, GBS, GAS, E. coli, S. aureus, C. difficile

Next Generation Technologies
- Structural Vaccinology
- Adjuvants/Human Immune Response
- Synthetic Biology
- Vectors

Vaccines For Every Age

**Pregnancy**
- CMV
- Flu
- GBS
- HBV
- Meningococcal
- Pertussis
- RSV
- Tetanus

**Infants & Children**
- Diphtheria
- Flu
- GAS
- HAV
- HBV
- Hib
- IPV
- Meningococcal
- Pertussis
- Pneumococcal
- Rotavirus
- RSV
- Tetanus
- Varicella
- Measles

**Adolescents**
- CMV
- DTaP boost
- EBV
- Flu
- HSV
- HPV
- Meningococcal

**Adults**
- Diphtheria
- Flu
- HBV
- Meningococcal
- Pertussis
- RSV
- Tetanus

**Elderly**
- Flu
- GBS
- Meningococcal
- Pneumococcal
- RSV
- Zoster
- Candida
- *C. difficile*
- *E. coli*
- *Klebsiella*
- *P. aeruginosa*
- Staph
- Breast Cancer
- Colorectal Cancer
- Prostate Cancer

# Vaccines For Today’s Society

<table>
<thead>
<tr>
<th>Poverty</th>
<th>Emerging Infections</th>
<th>Travelers</th>
<th>Patients with Chronic Diseases</th>
<th>Immunotherapy/Therapeutic Vaccines?</th>
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</thead>
<tbody>
<tr>
<td>Cholera</td>
<td>AIDS</td>
<td>Cholera</td>
<td>CMV</td>
<td>Cancer</td>
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<td>HAV</td>
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<tr>
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<td>JEV</td>
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<td>(HCV, HBV, HPV, HIV…)</td>
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<td>MenB</td>
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<td>Parasitic infections</td>
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<td>Typhoid fever</td>
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Discussion

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