Next Generation Multivalent PRINT® Nanoparticle Vaccine Targeting Pneumococcal Disease

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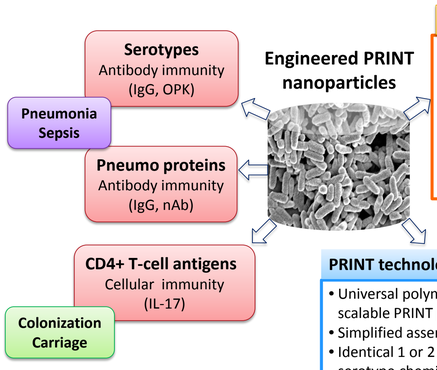
PRINT Technology Offers The Flexibility To Target Humoral And Cellular Immunity Towards Pneumococcal Specific Targets



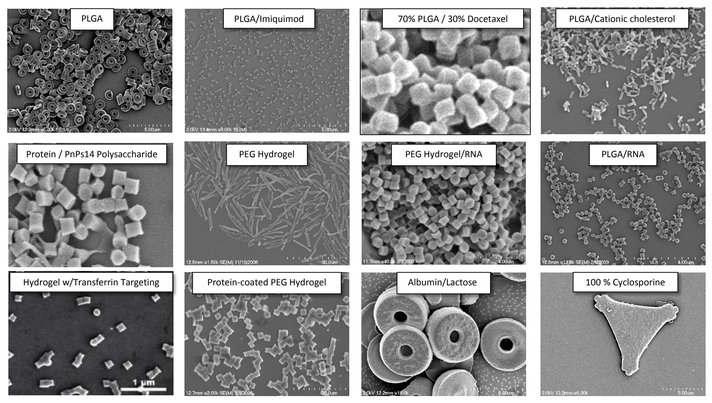
PRINT nanoparticle characteristics

- Enhanced B-cell/APC targeting and activation of T-cells
- Multiple copies of antigen incorporated
- Accessible to circulating antibodies
- Facilitates opsonization, neutralization
- Co-delivery of antigens, immunostimulants
- Effective primary, secondary immunization
- Improved delivery and efficacy

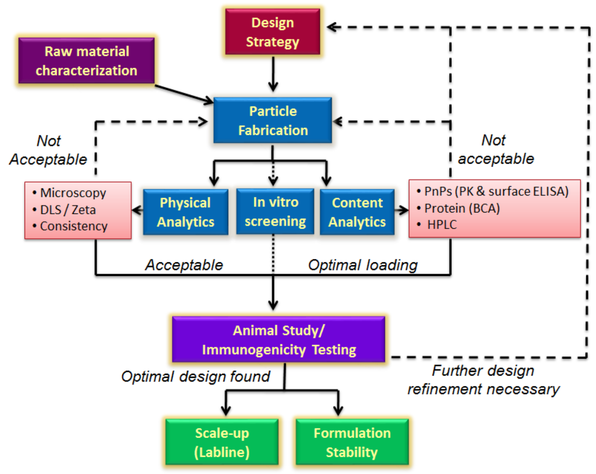
PRINT technology offers manufacturing advantages

- Universal polymer base manufactured by cost effective, scalable PRINT platform, well defined compositions
- Simplified assembly of multiple antigens
- Identical 1 or 2 step post-particle reaction, regardless of serotype chemistry
- No pre-treatment/activation of polysaccharide or protein
- Ease of purification from unreacted protein, polysaccharide
- Sterilization by filtration

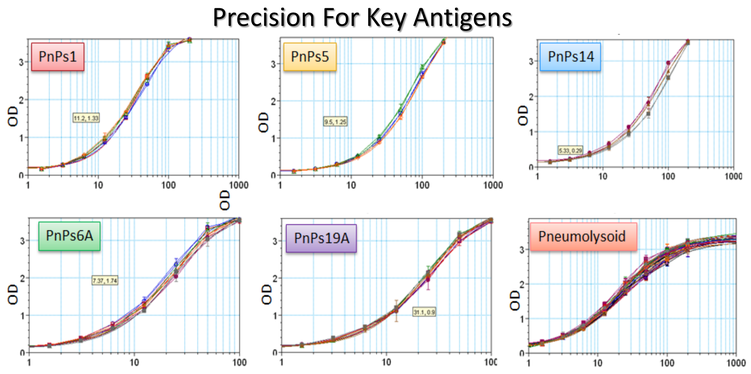
PRINT Is Compatible With Numerous Types Of Pharmaceutical Materials, Including Small Molecules And Biologics



PRINT Pneumococcal Polysaccharide Vaccine Development Pathway

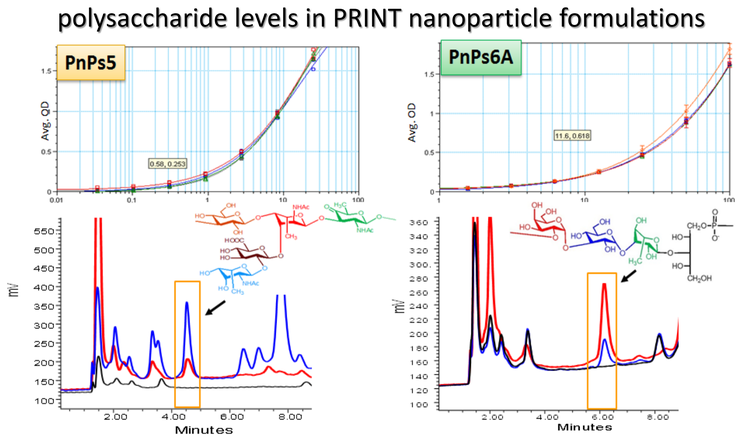


In-house IgG ELISA Shows Excellent Reproducibility And Precision For Key Antigens



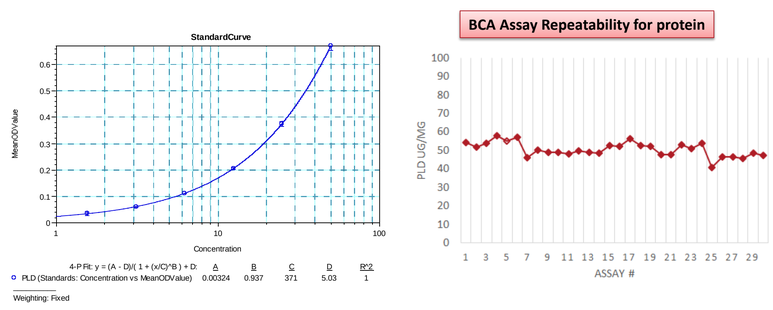
- The IgG ELISAs were adapted using standard W.H.O. IgG ELISA protocol
- PnPs specific IgG ELISAs have been adapted to all Prevnar13 serotypes
- Pneumococcal protein specific IgG ELISA also developed

In vitro ELISA and DIONEX HPLC methods to quantify polysaccharide levels in PRINT nanoparticle formulations



- Developed ELISA and DIONEX assays for all Prevnar13 serotypes

Protein in PRINT formulations can be reliably quantified in solid phase using a BCA colorimetric assay



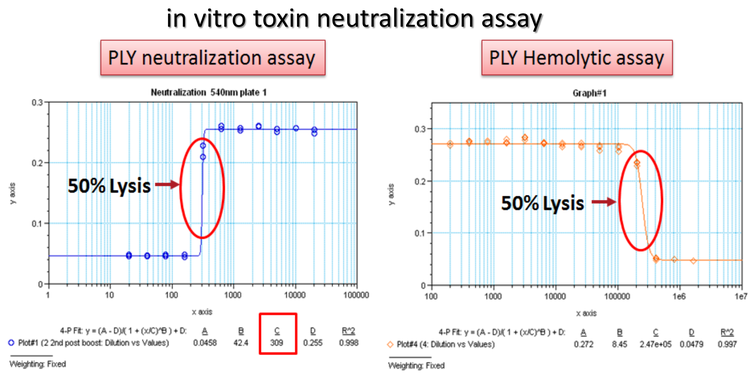
- Versatile method allows quantification of multiple antigenic carrier proteins in solid phase
- Good assay-to-assay precision for protein analysis in nanoparticles

Snapshot of Assays Developed for Multi-Serotype PRINT Pneumococcal Vaccine

Serotype/ Antigen	PnPs, Protein Detection in PRINT (ELISA, DIONEX, BCA)	IgG ELISA	*In Vivo* Evaluation (IgG, OPK, nAb)
1	✓	✓	mice, rabbits ✓
3	✓	✓	
4	✓	✓	mice ✓
5	✓	✓	mice, rabbits ✓
6A	✓	✓	mice ✓
6B	✓	✓	
7F	✓	✓	
9V	✓	✓	
14	✓	✓	mice, rabbits ✓
18C	✓	✓	
19A	✓	✓	mice ✓
19F	✓	✓	
23F	✓	✓	mice ✓
PLD	✓	✓	mice, rabbits ✓
PspA	✓	✓	mice ✓

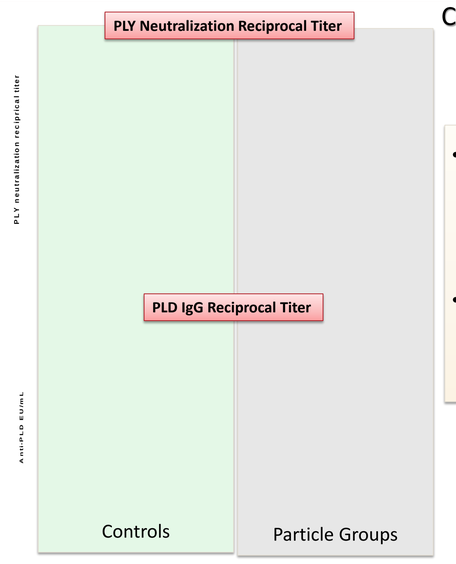
- ~9 Pneumococcal antigens successfully formulated with PRINT particles and evaluated in vivo to date
- Simplified and flexible assembly of antigens with PRINT particles translates easily into assay development

Functional Assessment of Pneumolysin mutant (PLD) in an in vitro toxin neutralization assay



- Neutralization assay developed to evaluate whether antibodies towards PLY have the capacity to neutralize hemolytic activity of the toxin
- Hemolysis inhibition (HI) assay of PLD was developed and harmonized between multiple sites/PATH collaborators

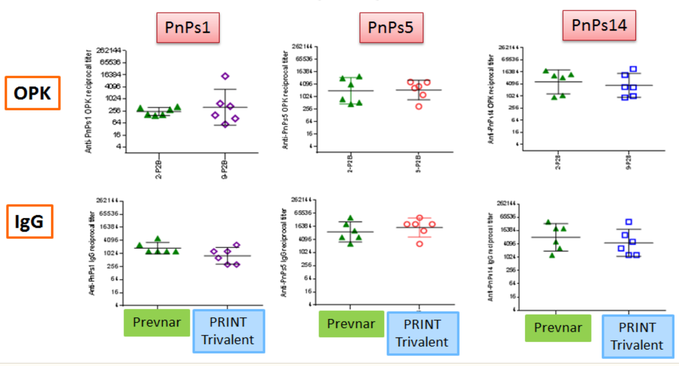
Correlation between PLY nAB and Anti-PLD IgG



- Robust PLD IgG antibodies were generated and shown to be functional by neutralizing hemolytic activity of PLY
- Strong correlation seen between PLY neutralization titers and PLD IgG ELISA Units/mL

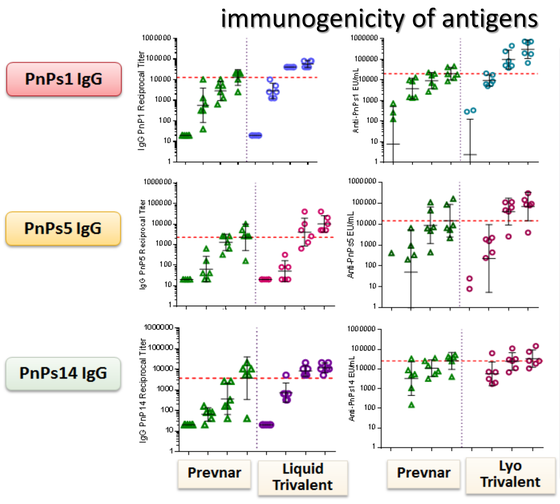
R^2= 0.831

Trivalent PRINT Pneumococcal Polysaccharide Vaccine Elicits Functional Antibody Response in Rabbits



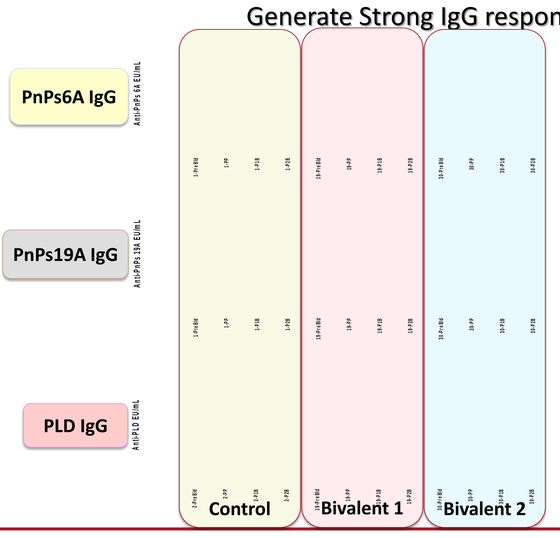
- Meaningful correlation between OPK and IgG observed in mice and rabbits
  - Functional antibody responses by opsonophagocytic assays (OPAs) is considered to be useful as a surrogate marker for protection
- Sera from mice vaccinated with licensed vaccine and PRINT formulations were analyzed by OPK assay in collaboration with Dr. Moon Nahm, University of Alabama

Lyophilization of PRINT formulations does not diminish the immunogenicity of antigens



- Lyophilized PRINT formulations induce significant immune response, equivalent to freshly prepared liquid formulation
- Lyophilization has the ability to allow for longer storage and minimal degradation of antigenic components and particle structure

Specific Bivalent Formulations of Additional Serotypes Generate Strong IgG responses



- Two non-adjuvented bivalent formulations showed statistically equivalent responses to Prevnar13 when tested individually for IgG response against each antigen
- PLD IgG responses were strong compared to soluble PLD alone

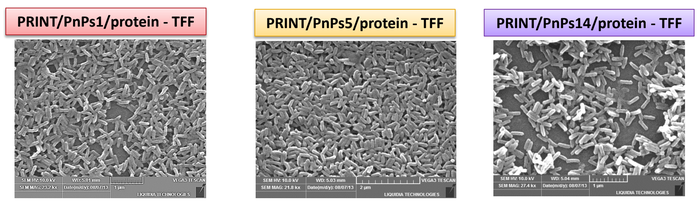
PRINT 80x80x320 nm vaccine formulations are sterile filterable

(80x320 nm PRINT particles used for Influenza Clinical Program)

Characteristics of filtered PRINT nanoparticle dispersions *(Early Trends)*	
PnPs loading efficiency *(serotype, process dependent)*	25 -50% *(optimization ongoing)*
Protein loading efficiency (serotype, process dependent)	~60-90% *Generally, high protein content observed*
Filtration Recovery Yield	> 65%
DLS (nm) / PDI	300-600 nm / 0.2-0.4
Surface charge	+10 to -15 mV *(design, process, serotype dependent)*

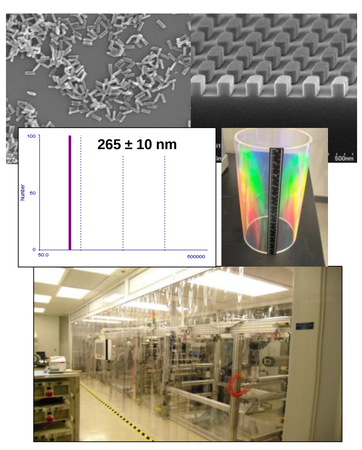
- PRINT technology enables process scale-up and allows for formulation activities to be integrated during TFF
- Common vaccine excipients used for filtration (polyvinyl alcohol, mannitol, sucrose, glucose, trehalose)
- Process parameters and scale-up activities currently ongoing

PRINT/PnPs1/protein - TFF | **PRINT/PnPs5/protein - TFF** | **PRINT/PnPs14/protein - TFF**



PRINT Manufacturing Advantages

- Quality by Design:
  - Unprecedented ability to independently control size, shape and composition
  - Uniform particle population, easily characterized
- Scalable Manufacturing Platform:
  - Proprietary tooling process enables translation of patterned Silicon wafer to patterned FCR mold
  - Leveraging the breadth of Roll-to-Roll technology - adapting, not inventing machine scaling
  - Commercially Relevant Scale and Cost
- Minimal Facilities Burden:
  - Small footprint, low CapEx equipment (potential for multiple, dedicated machines)
  - Low bioburden process, minimal water needs
  - No exotic utilities (drop-in-place equipment)
  - Concept for aseptic manufacturing, if required



PRINT platform demonstrates a wide-ranging multi-antigen formulation, manufacturing and analytical capability

- Robust analytical toolbox and infrastructure developed to quantify PRINT formulations antigen, protein content and IgG ELISAs (mice, rabbits) for all Prevnar13 serotypes
- Functional OPK assay test results strongly consistent with ELISA results
- Neutralizing antibody and IgG responses on par with soluble control; meaningful correlation between nAb and IgG trends
- Demonstrated the effectiveness of downstream sterile filterability and lyophilization of PRINT formulations
- The Print manufacturing process allows for rational design of next generation multivalent vaccines to address developing world needs.

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