

A Novel Nanoemulsion Adjuvant Enhancing The Immune Response from Intranasal Influenza Vaccine in Mice

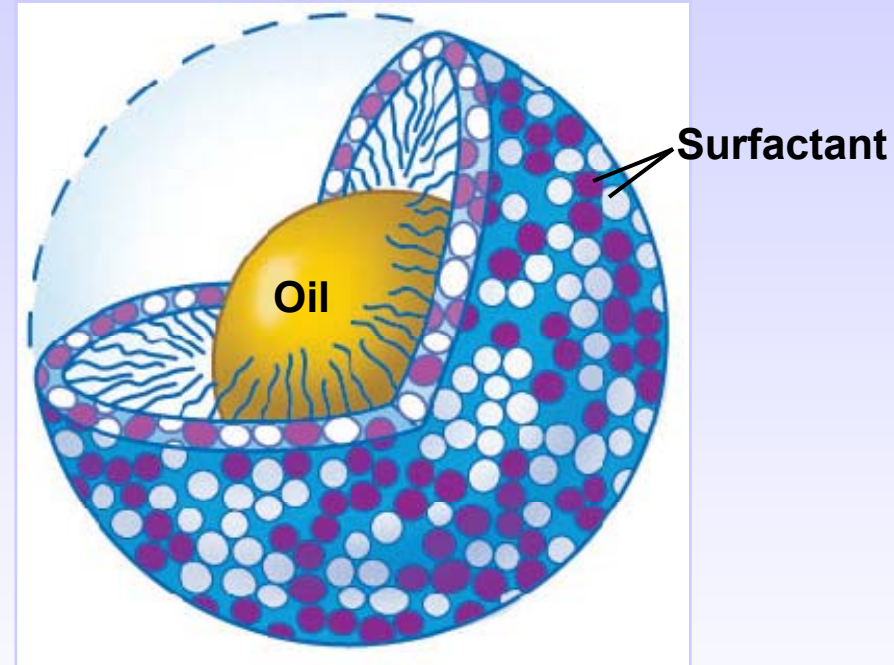
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Disclosure: Joyce Sutcliffe is VP, Research at NanoBio Corporation

What is a Nanoemulsion?

- High energy emulsification of:
 - Oil
 - Water
 - Organic Solvent
 - At least one surfactant
- Average Droplet Size
 - 400 nm

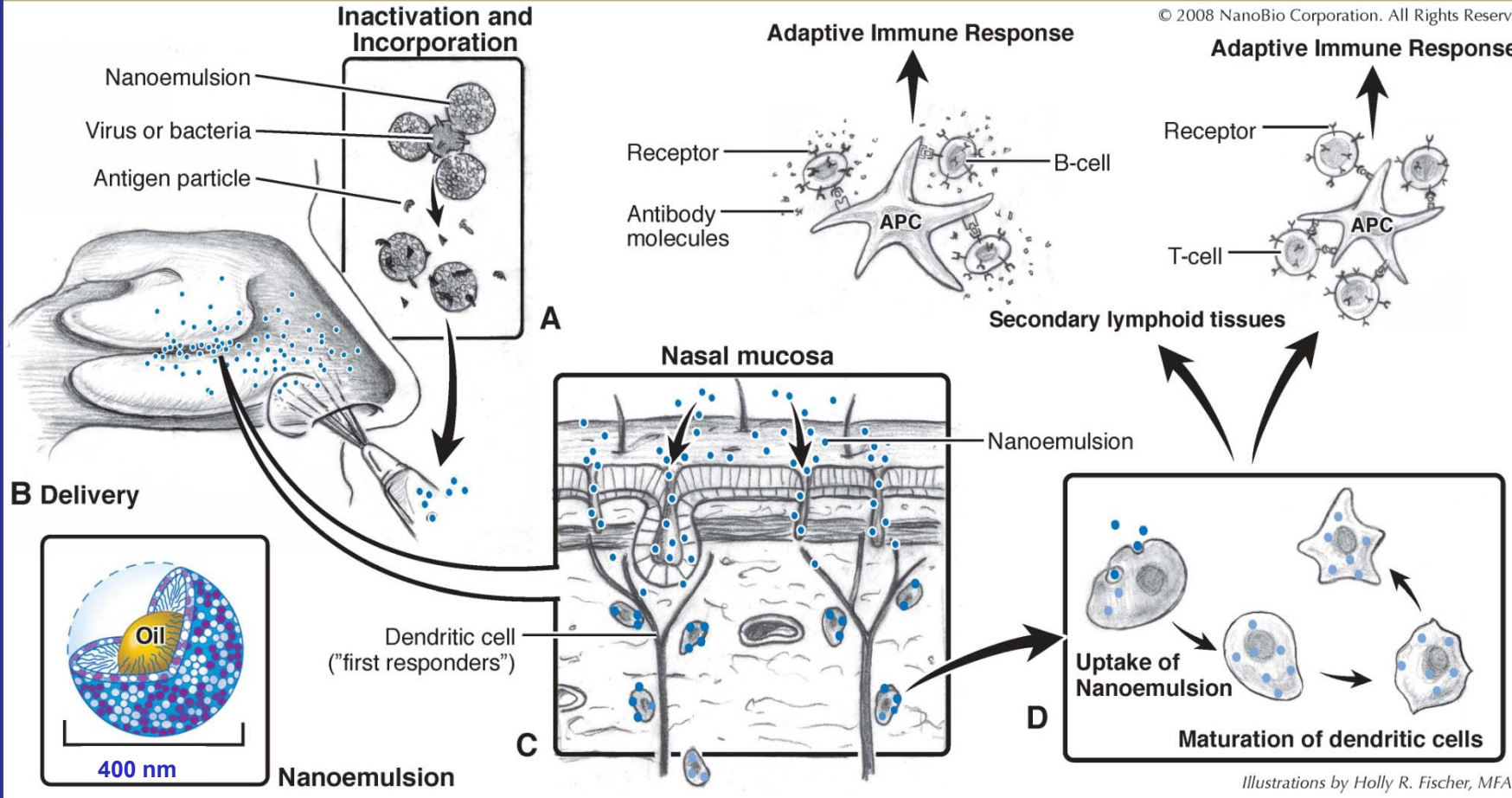


**Nanodroplet Itself is Integral
to Activity and Safety**

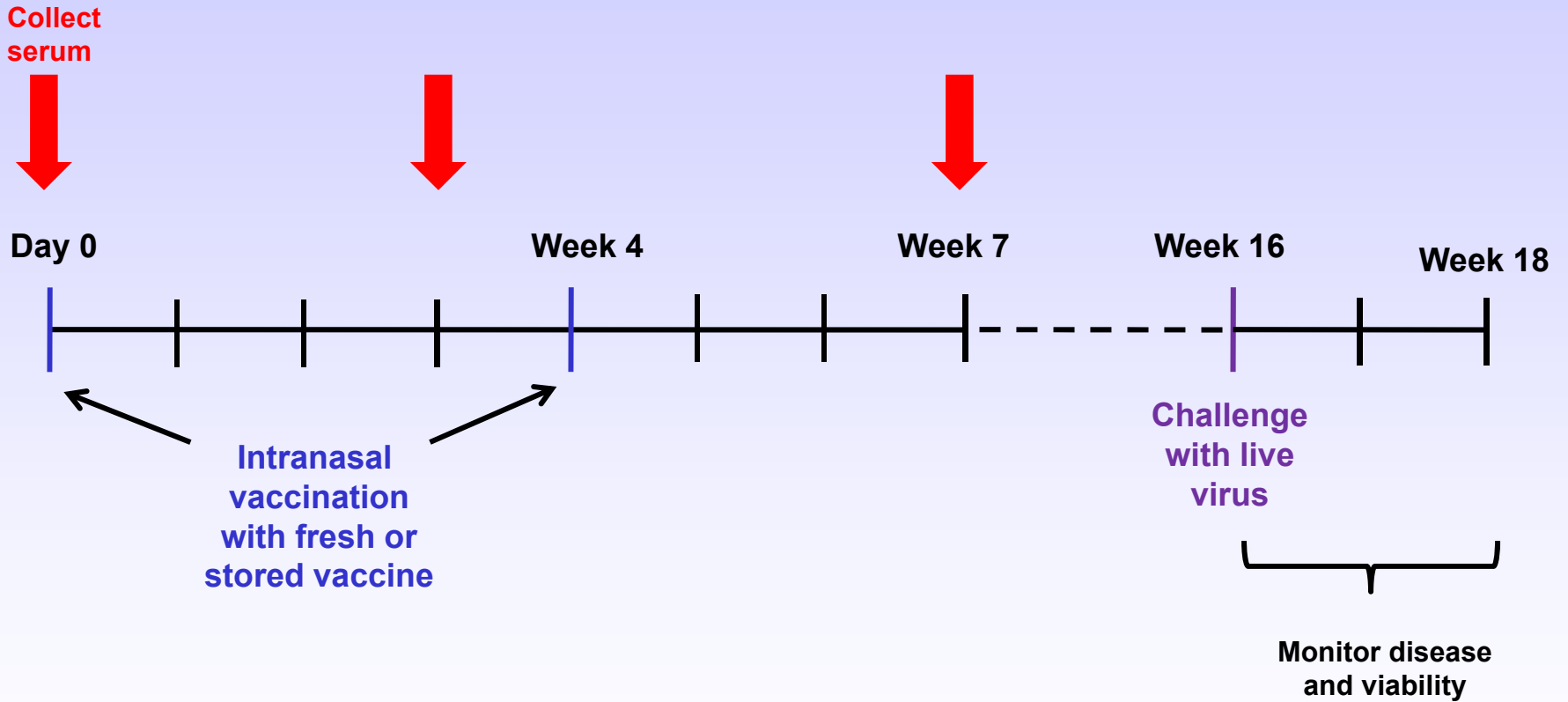
Mechanism of Nanoemulsion-Based Vaccine

MECHANISM OF ACTION IN NANOEMULSION-BASED VACCINES

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- **Determine the immune response and protection from subsequent challenge in CD-1 mice when given influenza A/Puerto Rico/8/34 (H1N1) ...**
 - Inactivated with nanoemulsion
 - Inactivated with β -propiolactone (β PL) and then mixed with nanoemulsion
- **Determine stability of nanoemulsion-based influenza vaccines over 3 months at either 4°C or 25°C**



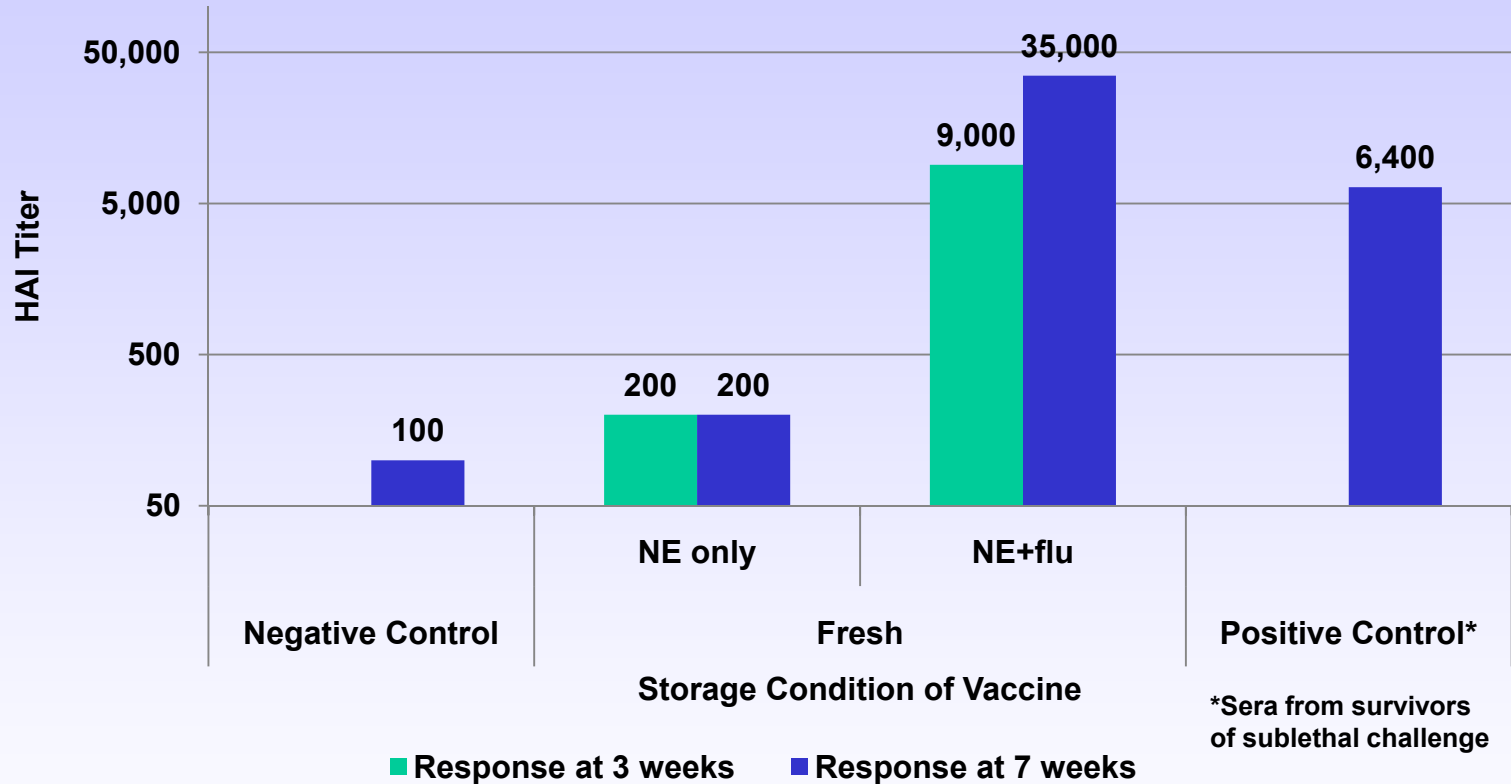
Inactivation of PR Influenza by Nanoemulsion

Virus Concentration (pfu)	Inactivation Time at 37°C				
	1 hr	2 hr	3 hr	8 hr	22 hr
10 ⁸	+*	+	+	-	-
10 ⁷	+	+	-	-	-
10 ⁶	+	+	-	-	-
10 ⁵	+	+	-	-	-

*presence (+) or absence (-) of CPE after 3 serial passages of incubation with MDCK cells

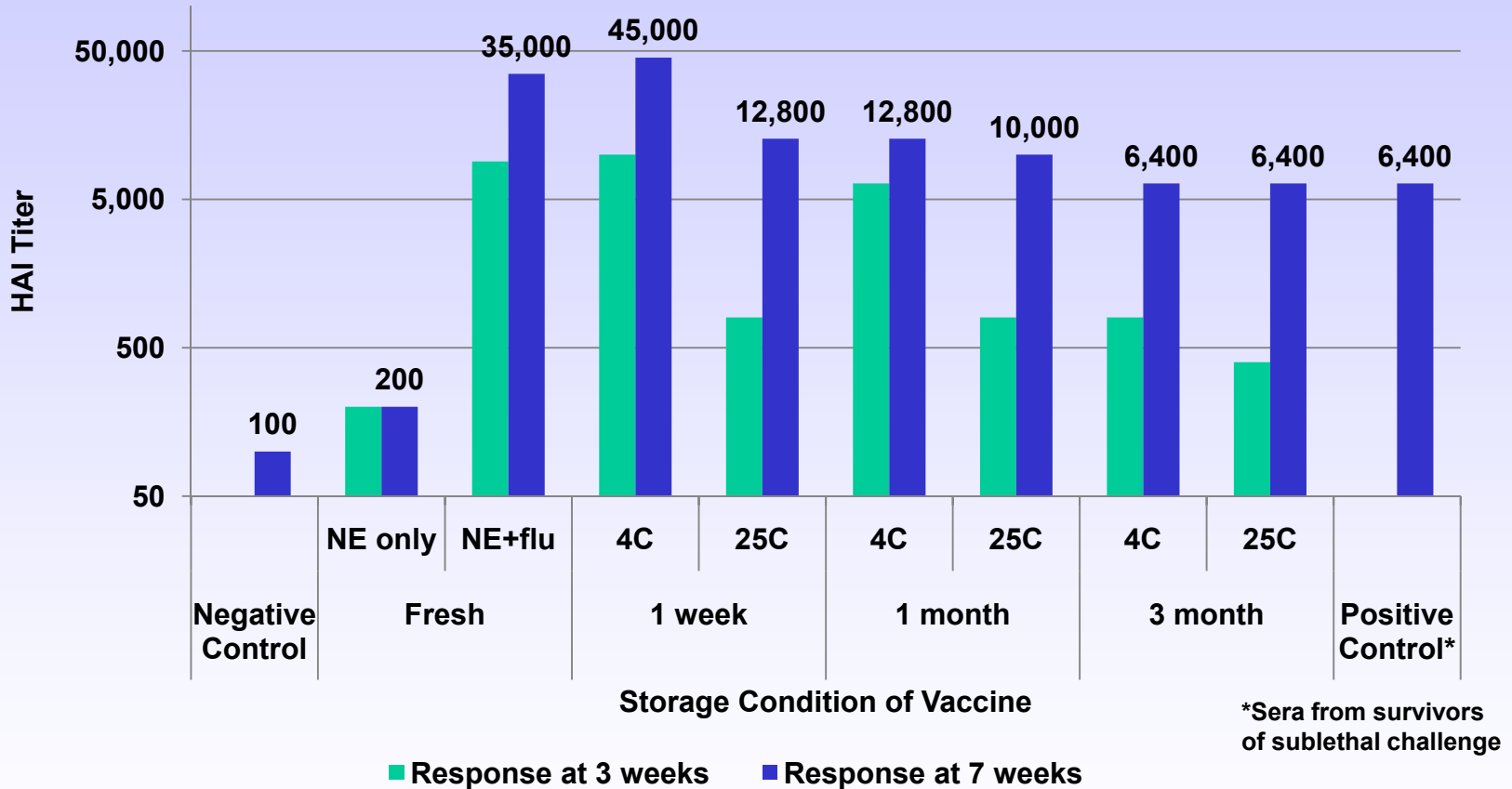
5 hrs at 37°C was used to inactivate 10⁷ pfu/ml of PR influenza virus

HAI Titers in Response to Nanoemulsion-Inactivated Influenza Vaccine



HAI Titers were as high as 90-fold or 350-fold over the negative control after a *single* or 2 vaccinations, respectively

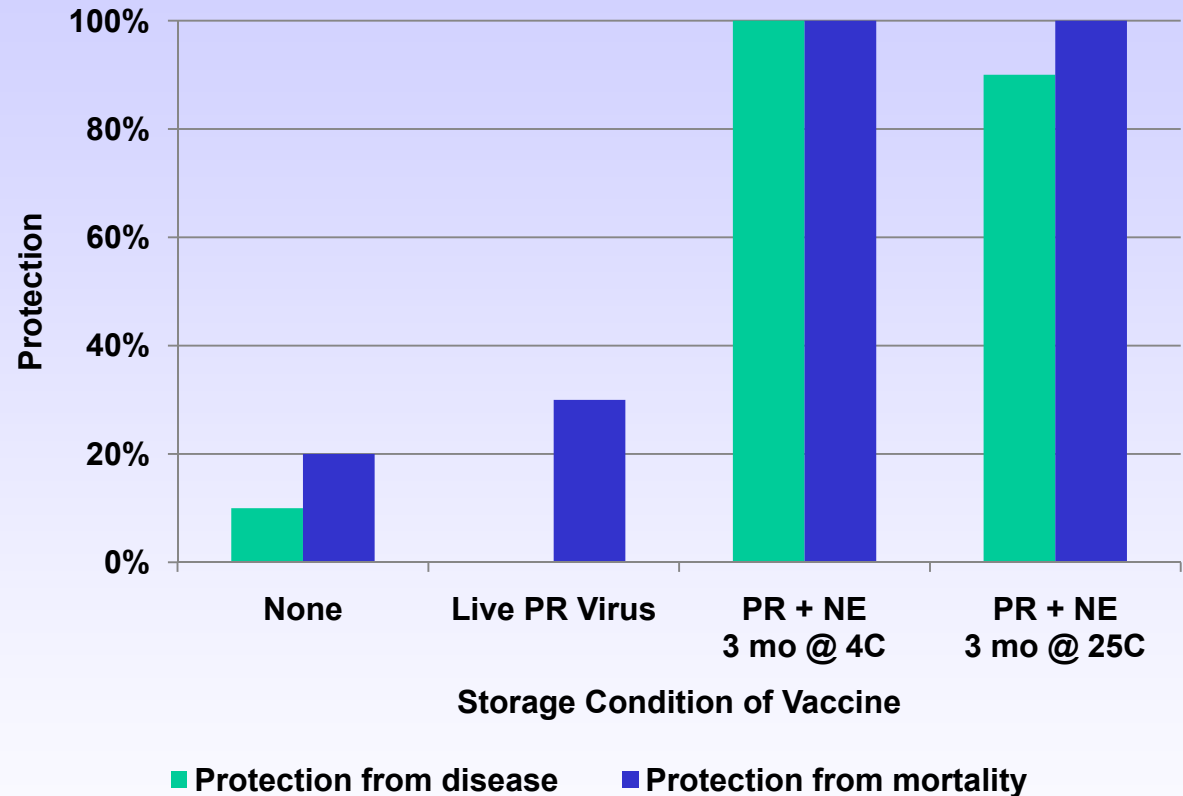
HAI Titers in Response to Nanoemulsion-Inactivated Influenza Vaccine



Vaccine stored at room temperature x 3 months elicited a strong immune response

Protection Conferred by NE-Inactivated Influenza Vaccine

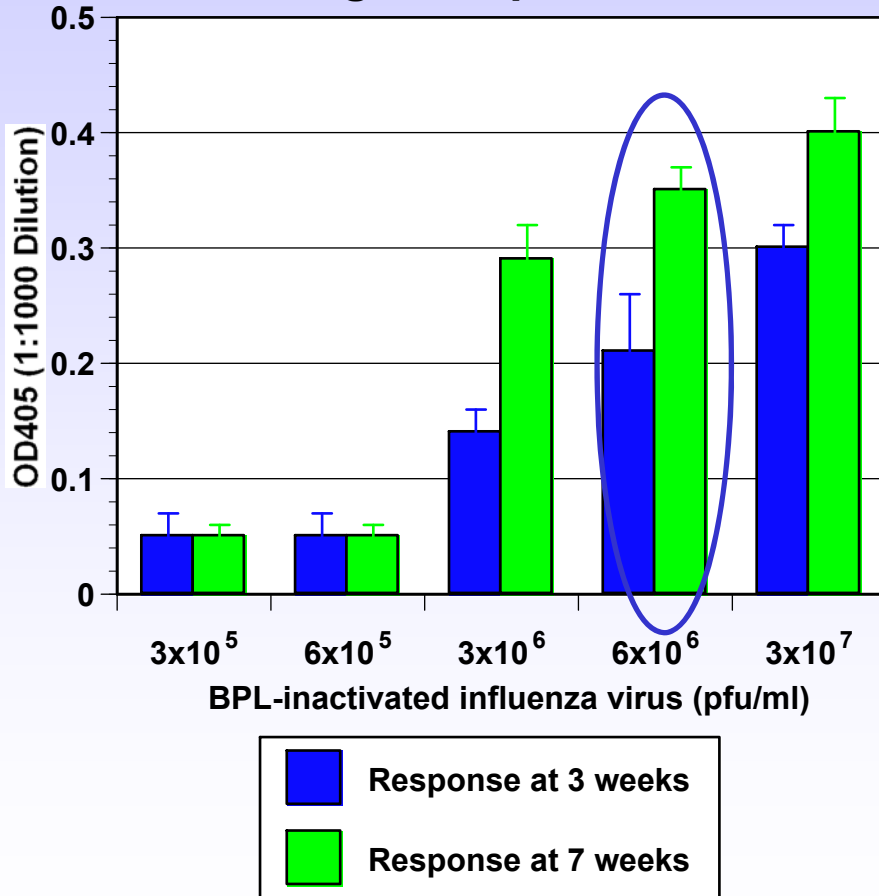
- After vaccination, mice were challenged with 100x LD₅₀ influenza virus (PR strain)
- Protection was evaluated for 14 days



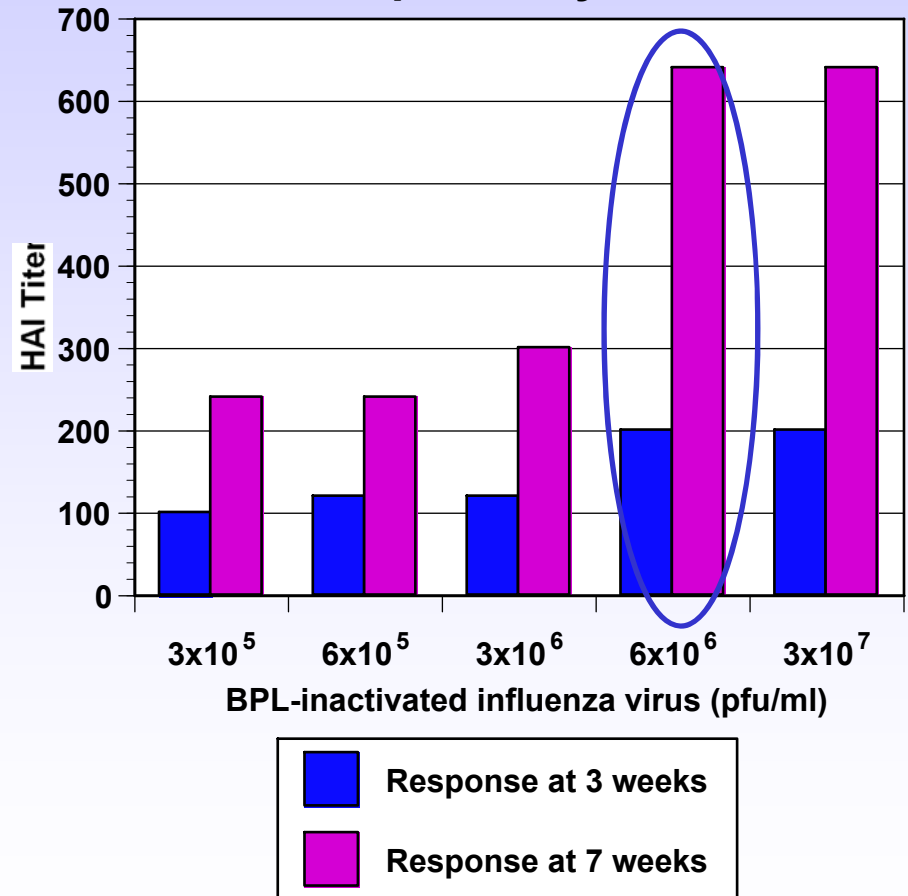
Virtually 100% protection from disease symptoms and death with either refrigerated vaccine or vaccine stored at 25°C for 3 months

Dose-Ranging: Immune Response to β PL-Inactivated Influenza Virus + Nanoemulsion

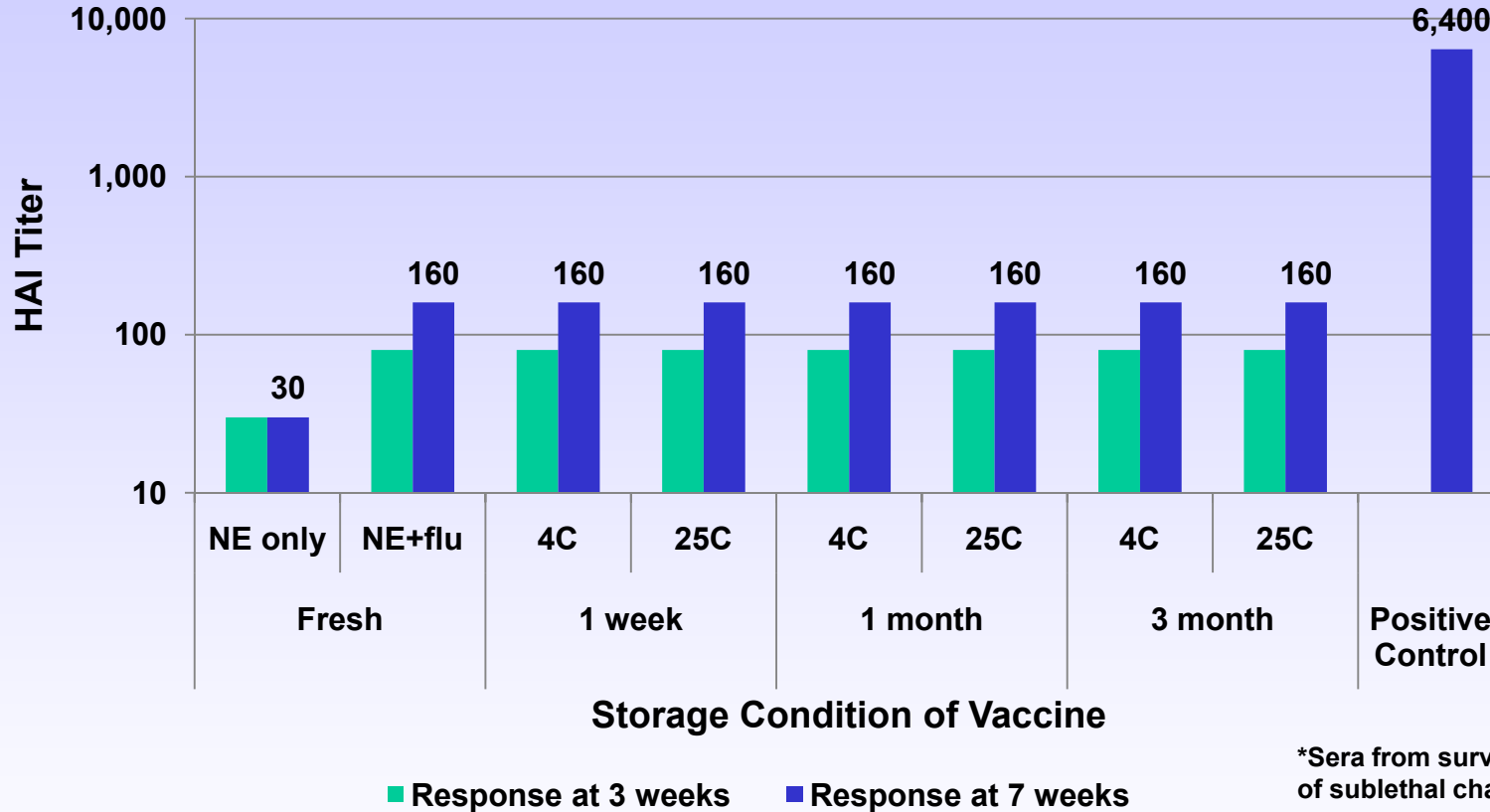
IgG Response



Response by HAI

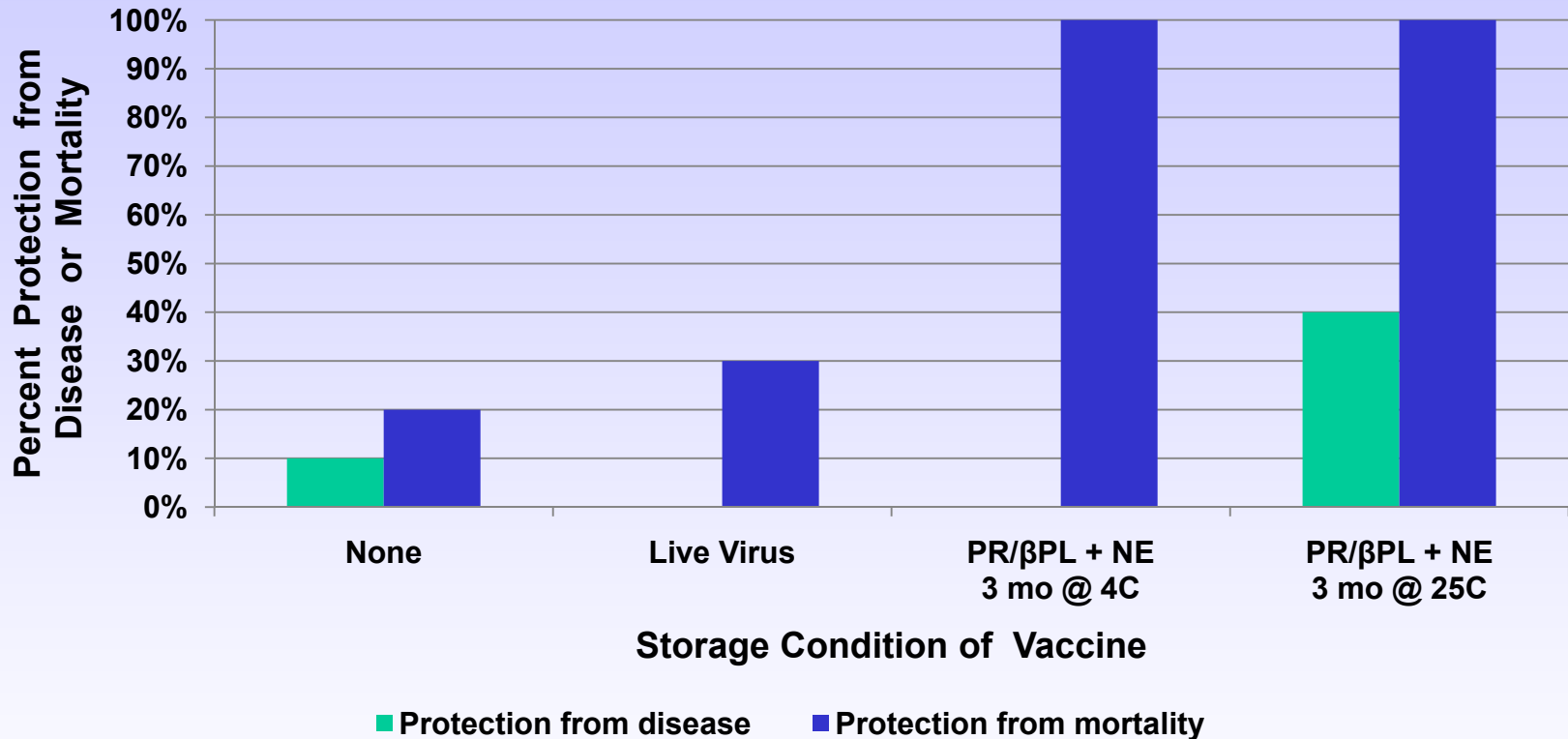


HAI Titers in Response to β PL-Inactivated + NE Influenza Vaccine



HAI titers were 5-fold higher for β PL-inactivated virus + nanoemulsion (\pm storage for 3 months) after 2 vaccinations

Protection Conferred When NE Mixed with β PL-Inactivated Influenza



Despite modest HAI titers, vaccine composed of β PL-inactivated influenza mixed with nanoemulsion protected against death, even after storage for 3 months at 4°C or 25°C

- **Nanoemulsion-based influenza vaccine**
 - Robust immune response after a *single* intranasal vaccination
 - Fully protected against challenge
 - Stable for 3 months at room temperature
- **Nanoemulsion mixed with β -propiolactone-inactivated virus**
 - Modest immune response after 2 intranasal vaccinations
 - Fully protected against mortality with subsequent challenge
 - Was stable for 3 months at room temperature

- **Nanoemulsion-based vaccines**
 - **Adjuvant the immune response**
 - **Elicit a strong protective response**
 - **Intranasal administration (needle-free)**
 - **Do not require cold storage**
 - **Safe**
 - **Work by a non-inflammatory mechanism**

Nanoemulsion-based vaccines provide advantages not equaled in current vaccines